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PRINCIPLES OF NATURAL COEXISTENCE INDICATED BY LEAFHOPPER POPULATIONS

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The generalization that no two species of organisms can occupy the same ecological niche at the same time has been expressed as Gause's Law (Lack, 1949), although there appears to be some doubt as to such an exact application of Gause's work (Gilbert, Reynoldson, and Hobart, 1952). In apparent contradiction to this generalization, we have at times found different insect species living in what appear to the collector to be identical situations. This has led us to seek sets of observations which would elucidate the problem. In particular, certain leafhoppers which feed on the eastern sycamore (*Platanus occidentalis*) seemed to offer evidence pertaining to this matter of ecological occupancy. A series of collections of these insects was made, and the data are here summarized. I believe that they permit significant conclusions indicating that certain of our concepts of species ranges and ecological occupancy have been unnatural.

THE SUBJECTS OF THE STUDY

The insects forming the subjects of this study comprise the *lawsoni* complex of the leafhopper genus *Erythroneura*. This complex contains six species, *arta* Beamer, *bella* McAtee, *lawsoni* Beamer, *morgani* DeLong, *torella* Beamer, and *usitata* Beamer. All six breed only on sycamore, and are the only leafhoppers that do so. All appear to have identical habits, the generations of the various species maturing synchronously in each locality, hibernating together, and feeding in the same manner, often side by side on the same leaf.

Two of the six species are readily diagnosed in both sexes by their color pattern: *morgani* has two black bands across the wings, and *bella* has a red dorsal saddle. The other four are whitish with red spots or bars; the males can be identified readily by characters of the internal genitalia, but the females cannot as yet be differentiated. Interspecific differences in male genitalia are not great, yet no intermediates have been found in several thousand specimens studied. Accordingly, I conclude that these distinctive morphological categories represent true species which hybridize little if at all. All six species also form a monophyletic unit closely allied to other species complexes which feed on maples, hazel, or other hosts.

Evidence from the geological history

of the host genus *Platanus* and from phylogenetic and dispersal evidence concerning the entire genus *Erythroneura* indicates that a population of *Erythroneura* became established on sycamore some time after mid-Tertiary, and evolved on this host into the *lawsoni* complex of six species.

The *lawsoni* complex occupies probably the greater part of the range of the eastern sycamore, which occurs throughout the eastern deciduous forest from Texas to Maine, figure 2. Outside of Illinois, few records of these leafhoppers are available. What records are available show that each of the six sycamore species is widespread; that *bella* is the most restricted, occupying the center of the sycamore range, with records from Maryland to the southern portion of Illinois; and that *lawsoni* is the most widespread, with records from Maryland to Kansas and Texas. To date no records of the complex are known from the New England area. Illinois therefore is a section of the composite range of all the species in the complex, which extends from the middle area where all six species are abundant, to practically the northern edge of the range of the complex.

This brief review shows that the Illinois populations of these sycamore leafhoppers represent a group of closely related species which (1) differ to a minimum degree genetically and therefore physiologically, (2) approach as closely as one could expect the status of ecological equivalents, and (3) appear to occupy the same niche (leaves of one species of plant) simultaneously throughout the same area. Furthermore, the Illinois populations appear to be representative of a reliable cross-section of the whole range of the species complex.

This investigation was aimed at answering these simple questions: How are these species distributed in the area with relation to each other? What general principles might be formulated to explain the distribution pattern?

Methods of Study

Collections of these leafhoppers were made over most of Illinois. An effort was made to obtain a sample of at least 100 specimens from each tree or grove visited, but in some cases leafhoppers were so scarce that only small numbers could be secured. Collections were obtained in a variety of situations and at more or less uniformly distributed points over all of Illinois. Complete uniformity of sampling was difficult to achieve because of the sparseness of sycamore trees in many areas of the state, or because of the inaccessibility for sweeping purposes of trees in certain situations. In some areas it was necessary to use samples from trees planted in towns. In all, 138 collections were secured, representing 87 of the 102 counties in Illinois. These collections were taken chiefly in 1953, 1954, and 1955. Inasmuch as these leafhoppers appear on the host plants simultaneously and occupy the host for about the same length of time, a seasonal replacement or similar variation in species composition during any one summer is not to be expected. Accordingly, the annual collection provides a reliable indication of the niche inhabitants, and it is unnecessary to analyze the collections by months or weeks.

Each collection was identified, and the percentages of each species were calculated. The order of abundance of each species (first, second, third, etc.) was then tabulated and plotted on maps of Illinois.

Relative General Abundance

One of the first analyses of the sycamore collections was undertaken in an attempt to find some measure of the total relative abundance of the various species. Histograms were made based on the number of times each species was first, second, third, and so on, in the various collections, figure 1. These were first plotted in 1954, including chiefly collections made in 1953 and 1954, with a few sporadic

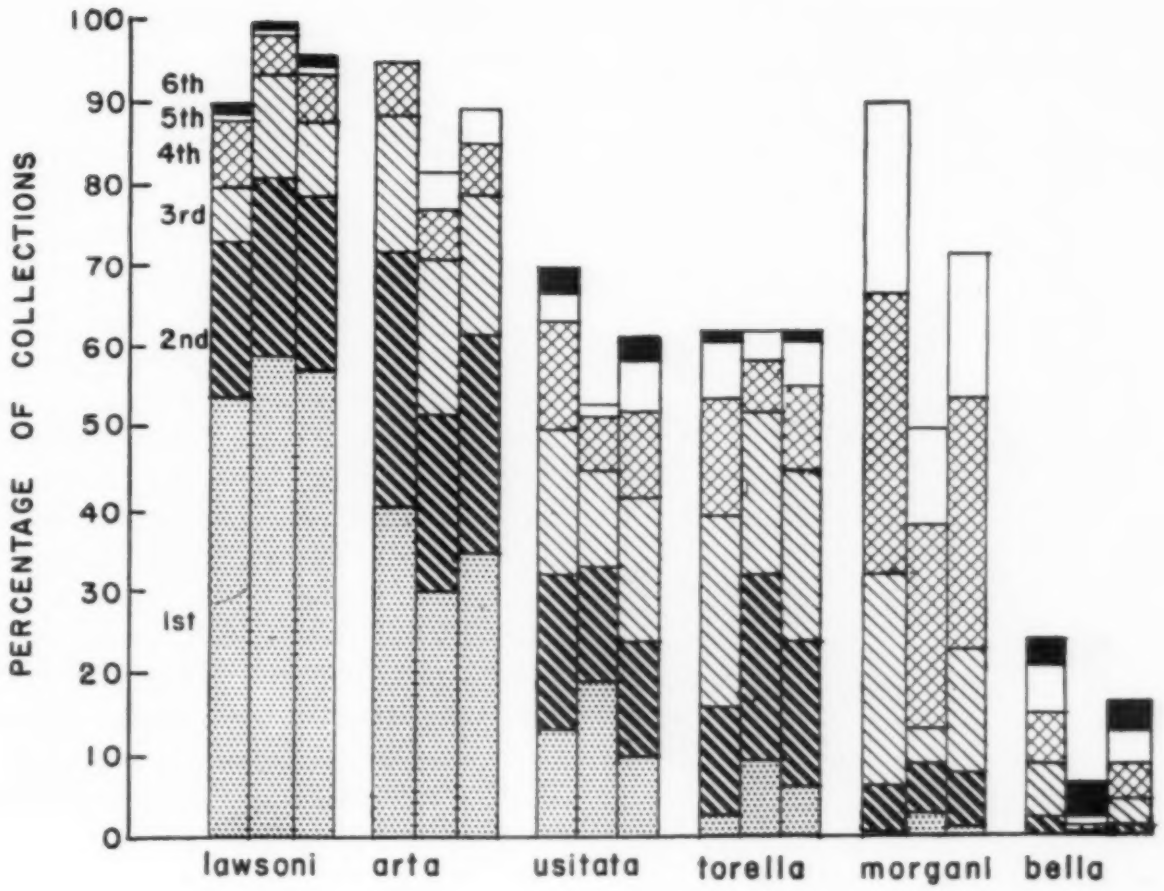

FIG. 1. Rankings of sycamore leafhopper collections in Illinois. In the tabulation of each species, the left column represents collections made prior to and during 1954; the center column, collections made in 1955; the right column, total of all collections.

collections made in previous years. This summary contained two weaknesses. It mixed collections from several years, and it included no collections from a large section of northwestern Illinois. More extensive sets of collections were made in 1955 in order to assemble distribution data from the neglected area and to obtain some measure of year to year fluctuations. In figure 1 data up to and including 1954 are given in the first column for each species, 1955 data are given in the second column, and the average for all collections is given in the third column. Because it includes no collections from extreme southern Illinois, the middle column has a decided bias in favor of *lawsoni*, the most abundant northern species, and against the more southern species such as *bella* and *usitata*. When allowance is made for this bias, there is good general agreement between the columns of a species.

It is readily evident that *lawsoni* is the predominant species, *arta* is second in abundance, *usitata* and *torella* are fairly evenly matched for third and fourth, *morgani* is fifth but does have a surprising number of collections to its credit, and *bella* is very low on the scale of relative abundance.

Remembering that these species live together on the same trees, one is tempted to ask at this point: Why does not the most abundant species swamp out or crowd out the less abundant species, or at least the lowliest one, *bella*, which is out-produced by all five other species on sycamore?

GEOGRAPHIC DISTRIBUTION AND RELATIVE ABUNDANCE

Two species, *lawsoni* and *arta*, occur over the entire length of Illinois. Three species, *morgani*, *usitata*, and *torella*, have been collected only in the southern four-fifths of the state. The sixth species, *bella*, has been found only in the southern

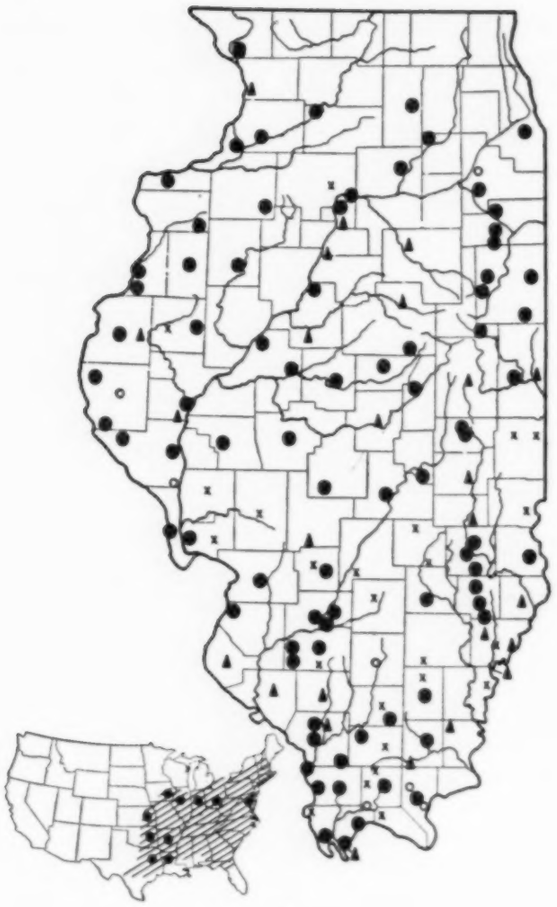

FIG. 2. Relative abundance of *Erythroneura lawsoni* in Illinois sycamore collections. On the Illinois map, large solid circles represent first rankings; black triangles, second rankings; crosses, lower rankings; and small hollow circles, negative collections. On the inset map, the hatching indicates the range of sycamore, the dots, state records of *lawsoni*. These symbols are used also in figures 3 to 7.

three-fifths of the state. In the southern three-fifths of the state, therefore, all six sycamore species are found, in the next fifth only five species, and in the northern fifth only two species. I must confess that, knowing this in general before the project started, I fully expected to find that the more northerly species would predominate toward the north and the more southerly species would predominate toward the south. This idea proved to be a gross over-simplification, as the following account of each species will show.

Lawsoni, figure 2. This was the most abundant species throughout Illinois except in the southern seven counties, where *arta* equalled it in number of first places. Of the 138 collections made, *lawsoni* was absent from only six, and ranked first in 78. Over the southern four-fifths of the state, the six negative collections, the first rankings, and the lower rankings were scattered irregularly.

Arta, figure 3. This was the second most abundant species, occurring over the full length of the state. In contrast with *lawsoni,* the 20 negative collections of *arta* are chiefly from the northern half of the state. *Arta* had only 49 first rankings. These first rankings equalled those of *lawsoni* in the southern part of Illinois, but became fewer and more scattered northward. In spite of this general relative decrease in abundance northward, *arta* was the predominant species in some collections practically at the northern extremity of its range.

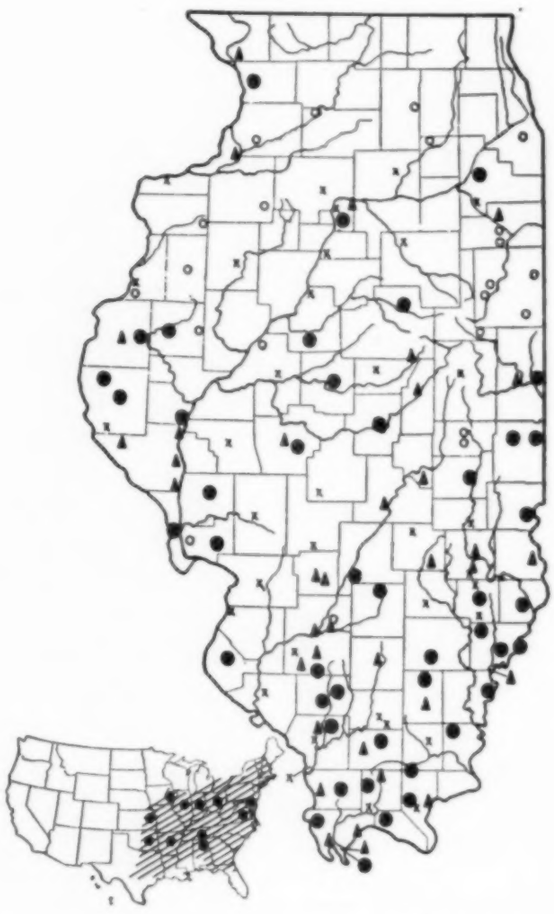

FIG. 3. Relative abundance of *Erythroneura arta* in Illinois.

Usitata, figure 4. Compared with *lawsoni* and *arta,* this species has a low general abundance. It has only 14 first rankings and was absent in 51 collections. In the southern and southeastern areas of Illinois, however, *usitata* has few negative collections and a moderate scattering of first and second rankings. Northward the negative collections increase rapidly as the species approaches the northern limit of its range. In central Illinois *usitata* has many negative collections, but among them are scattered a few first and second place collections. The remarkable fact is that in nearly the most northern point at which we have collected it, this species ranks first in the collection. In the two most northern localities in which it has been found (only five miles north of the preceding), it was second in abundance, in both cases outranking *arta.*

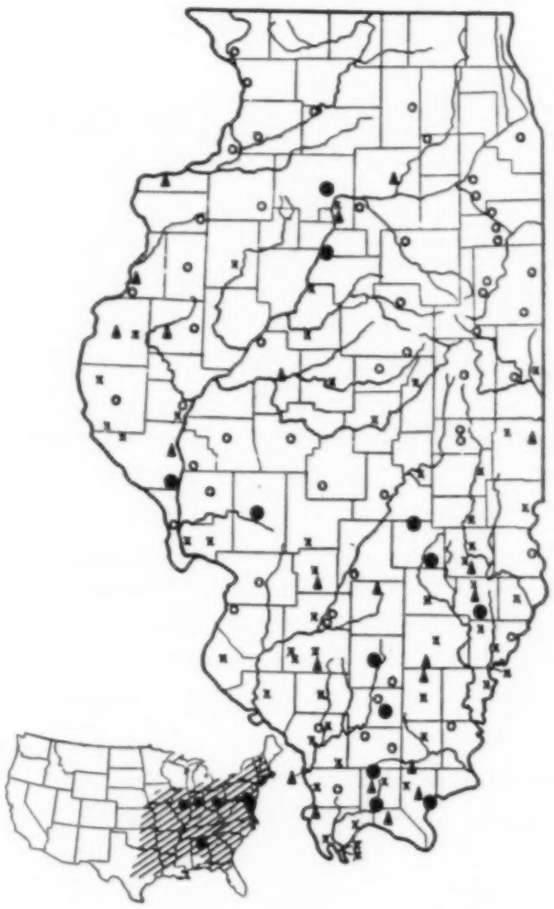

FIG. 4. Relative abundance of *Erythroneura usitata* in Illinois.

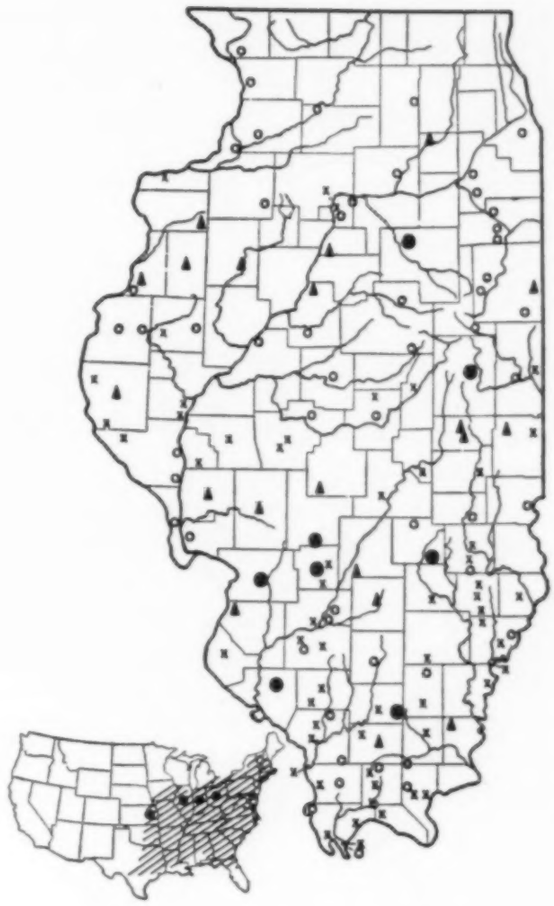

FIG. 5. Relative abundance of *Erythroneura torella* in Illinois.

Torella, figure 5. Here again we are dealing with a species of low general abundance, even lower than *usitata.* Its 50 negative collections are scattered from south to north. Its first rankings are most numerous in the second fifth of the state northward. As with *usitata,* however, some first and second place collections of *torella* occur along the extreme northern periphery of its range.

Morgani, figure 6. The outstanding feature of this species is the unusually large number of collections of which it comprises little more than a trace. In 24 of the 85 collections in which it occurred, *morgani* represents 3 per cent or less of the population sample. In total collections *morgani* slightly outnumbers both *usitata* and *torella,* but has only 3 first rankings and 7 seconds. Its total range in Illinois is almost coincidental with these two species. And as with

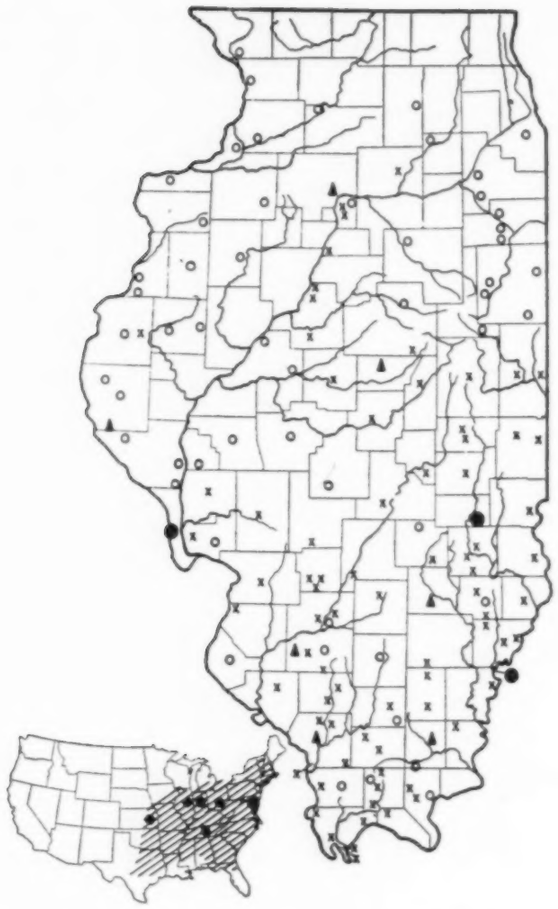

FIG. 6. Relative abundance of *Erythroneura morgani* in Illinois.

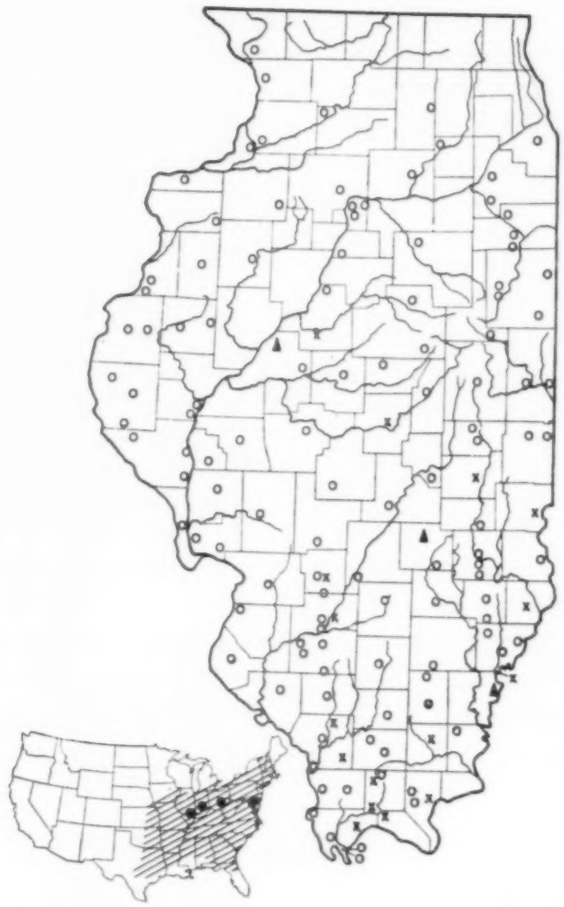

FIG. 7. Relative abundance of *Erythroneura bella* in Illinois.

torella and *usitata, morgani* forms a relatively abundant constituent of the population (second at 20 per cent) in one of its most northern localities.

Bella, figure 7. In Illinois this species is restricted to the southern three-fifths of the state. In general abundance it is the lowest of the six species in the complex. Only in extreme southern Illinois does it occur in a moderate proportion of the sycamore collections. In our total series it has 117 negative collections and no first rankings. It has a few second and third rankings, and these are scattered irregularly over the entire Illinois range, including one of the two peripheral northern records.

Inasmuch as the data for these six species were collected over a span of several years, the possibility arose that results from a single year would be markedly different. In an effort to check this point, the results for 1955 were plotted on one map, those for the total of other years on a separate map. In the case of each species the two maps were in agreement on the points listed above.

The data presented in these maps show that there is at most only a vague correlation between geographic location within the range and the relative abundance of each species. While each leafhopper species tends to be more abundant in one region and less so in others, this is not expressed in an evenly distributed decrease from an area of high density to one of low density. Instead, the proportions of each species vary locally to an extreme degree. A species may have a high density in a locality situated in an area of low average density, or a low density in a locality situated in an area of high density.

Ecological Distribution

Different sycamore trees may exhibit a wide variety of ecological conditions. The sycamore is primarily a floodplain species, yet it occurs also in some upland situations and at the heads of gullies supporting only ephemeral streams. Large numbers of sycamores have been planted along roadsides and in towns in a variety of situations. An effort was made to record observations which would be useful in correlating ecological characteristics of trees with the relative abundance of leafhopper species. A few useful correlations were found, but except to the north it was found impossible to predict the composition of the leafhopper fauna of a given tree from a gross ecological evaluation of the situation in which the tree occurred.

To test the human eye as a judge of similarity of ecological conditions of sycamore trees, a set of double collections were made in several localities. Each set was composed of two trees of about equal size which appeared identical ecologically in regard to shelter and humidity of the immediate surroundings. Data for three of these pairs are given in table 1. The two trees (A and B) at Versailles, Illinois, were separated by about half a mile, the two at Plano by 50 yards, and the two at Greenville by about 100 yards. The leafhopper samples on the two trees at Plano were remarkably similar; the other two pairs differed markedly with respect to some of the constituent species, little with respect to others.

The only species collected consistently on trees in open, dry, wind-swept situations was *lawsoni*. In many such collections it was the only species encountered. On the other hand, *lawsoni* was frequently the first-ranking species on trees in sheltered, humid valleys. This species thus has an ecological tolerance encompassing practically every situation in which sycamore trees normally grow. The species *arta* was found on trees in almost as great a variety of situations, but was seldom the first-ranking species in the more extreme open or dry situations.

On the opposite end of the scale, *morgani* and *bella* were seldom found except on trees in humid valleys or in well-protected situations. These species apparently require the most circumscribed conditions of all to develop into more than trace species.

The other two species, *usitata* and *torella*, were also found in at least fairly humid situations. Aside from this no correlation was evident between their abundance and the ecological conditions in which the trees were found.

From these observations it appears that all species of the *lawsoni* complex thrive under conditions of high humidity and shelter from evaporation, and that fewer species survive as the humidity or shelter

Table 1. *Leafhopper populations collected on duplicate trees A and B in the same locality and at the same time.* The first number refers to rank in the population, the second number to the actual percentage of the population.

		lawsoni	*arta*	*usitata*	*torella*	*morgani*	*bella*
Versailles	A	2–33	1–47	4–6	3–14	0	0
Versailles	B	1–55	2–29	0	3–16	0	0
Plano	A	1–80	0	0	2–20	0	0
Plano	B	1–78	0	0	2–22	0	0
Greenville	A	3–18	2–31	4–10	1–36	5–5	0
Greenville	B	1–36	2–18	2–18	4–15	5–11	6–2

TABLE 2. *Collections of* Erythroneura *on identical sycamore trees in successive years.* The first collection number refers to rank of the species in the population, the second number to the actual percentage of the population represented by each species. Differences between the two collections are indicated in the third line for each locality.

		lawsoni	*arta*	*usitata*	*torella*	*morgani*	*bella*
Camargo	1954	1–73	0	0	2–26	3–1	0
	1955	1–56	0	0	2–41	3–3	0
		0–17			0–15	0–2	
E. Peoria	1953	1–65	3–11	5–2	2–13	4–9	0
	1955	1–53	3–13	2–18	5–4	4–12	0
		0–12	0–2	3–16	3–9	0–3	
Hennepin	1953	2–27	1–48	3–21	4–3	5–1	0
	1955	1–41	3–11	2–37	0	4–11	0
		1–14	2–37	1–16	1–3	1–10	
Mounds	1950	2–23	1–65	3–6	0	3–6	
	1953	1–34	2–25	3–19	4–16	5–6	0
		1–11	1–40	0–13	1–16	2–0	
Murphysboro	1953	1–31	2–29	0	4–8	3–24	4–8
	1955	2–19	1–56	3–9	0	4–8	4–8
		1–12	1–27	3–9	1–8	1–16	0–0
Newton	1953	1–51	2–22	4–8	5–5	3–13	0
	1955	1–50	2–17	3–14	5–6	4–13	0
		0–1	0–5	1–6	0–1	1–0	

decreases, until chiefly *lawsoni* occurs at the extreme edge of the group's tolerance for these factors.

CHANGE WITH TIME

Collections were made on the same trees in two different years to obtain some measure of the changes occurring in population proportions with time. In six of these sets, ecological conditions appeared similar at the time of both collections. A perusal of these six sets of data, listed in table 2, indicates that the same type of population differences occur from year to year on the same tree as were noted between different trees in the same vicinity at the same time (see table 1). In very few cases are the percentages between paired collections identical for any species. In the Camargo and Newton sets, there was little or no change in standings, but the percentage of *lawsoni* dropped appreciably in 1955 compared to 1954. The two East Peoria collections demonstrate a reversal of position between *usitata* and *torella,* which is surprising in view of the relative stability of the rest of the population. In the Hennepin, Mounds, and Murphysboro collections the relative positions of four or five species changed. At Hennepin and Murphysboro, the position of *lawsoni* shifted from second to first, and from first to second, respectively. In the Murphysboro collection a simple first-second switch occurred between *lawsoni* and *arta,* whereas in the Hennepin collection *arta* dropped from first in 1953 to a poor third behind *lawsoni* and *usitata.*

These collections indicate the occurrence of population fluctuations from year to year which are variable in different localities both as to degree of change and the species involved.

Applying these results to the maps in figures 2 to 7, it is obvious that there is represented here only a cross-section of a highly plastic series of populations. It follows that maps based on data collected in other years would differ from the present ones in details concerning a given locality. It is probable, however, that taken as a whole these data would give the same over-all proportions as shown in figure 1.

Explanation of Results

The foregoing analysis of these sycamore populations demonstrates that the proportions of the various species differ in an apparently haphazard fashion (1) from place to place and (2) from year to year. In an attempt to explain such variability, three factors would normally be supposed to be operating: interspecific competition, vagility (ability to disperse), and ecological adjustment.

Competition

The data accumulated on the six species of the *lawsoni* group indicate that interspecific competition was not a factor affecting either their abundance or distribution during the period when these collections were made. Due to differences in collecting efficiency depending on wind velocity and temperature, and the difficulty in obtaining large samples from tall trees or those crowded in with other tree species, it is virtually impossible to devise a good method for determining the absolute abundance of leafhoppers per unit tree or unit area. It is easy, however, to estimate populations as small (roughly 1 to 50 leafhoppers obtained per medium-sized tree), medium (roughly 50 to 500 per tree), and large (over 500 per tree, often reaching many thousands); these are the categories used here. In this section I refer also only to the southern half of Illinois, where all six species are found as a coexisting complex.

If interspecific competition were a factor influencing the relative abundance of the sycamore leafhoppers, we would expect trees with small populations, where competition would be at a minimum, to have the greatest average diversity of leafhopper species. Conversely we would expect trees with the largest leafhopper populations, where competition would be greatest, to have the least diversity of species. But this is not what we find. Regardless of size or situation, trees with small populations usually harbor only *lawsoni* or *arta*, whereas trees with large populations almost always support all six species. I am therefore led to believe that interspecific competition is not a factor in regulating populations of these species.

This same phenomenon is illustrated strikingly by some other groups of *Erythroneura*. The shingle oak (*Quercus imbricaria*) has a large and restricted fauna of about a dozen species of *Erythroneura*. In most localities we have visited, it is difficult to obtain more than a dozen or two specimens from a clump of shingle oaks. Each collection usually has only two or three species. Certainly there is no competition between leafhoppers here, because a single small leafhopper may have a whole tree to itself. On one occasion we encountered a shingle oak which was "loaded," having many hundreds of leafhoppers; this collection contained every species ever recorded from the host. Similar examples could be cited for several other hosts on which we have collection records.

What appears to be interspecific competition does occur occasionally between these leafhoppers and the sycamore tingid or lacebug, *Corythuca ciliata* Say, which is also widespread on sycamores in Illinois. In 1955 in certain Illinois localities (Oregon and Kankakee), the tingids reached such a high population level that the sycamore leaves became coppery and brittle. *Erythroneura* were either absent or very scarce on such trees, many of

them so situated that they should have had a good *Erythroneura* fauna. It is likely that the excessive tingid feeding simply dried the leaves to such an extent that the *Erythroneura* could not get sufficient food out of them, and either left the host tree or died. We have too few data to know if this tingid action has a differential effect on different species of *Erythroneura.*

In contrast to the sycamore collections reported here, instances are known in which *Erythroneura* have completely etiolated their host. I have seen redbud trees (*Cercis canadensis*) in this condition and also on one occasion in Columbus, Ohio, a series of etiolated sycamore trees. These latter sustained immense numbers of *Erythroneura lawsoni.* Cultivated Virginia creeper and grape may be so severely etiolated by various species of *Erythroneura* that the leaves wither and fall in midseason. Under these conditions some sort of competitive effect or differential species survival may occur. That such conditions occur very rarely on sycamore, however, is shown by the fact that in ten years of collecting I have personally encountered only the single example of etiolation cited.

It seems certain, therefore, that if some form of interspecific competition occurs between the sycamore leafhoppers, it is extremely local and/or infrequent.

Vagility

The effect of vagility on *Erythroneura* populations is obscure. Collections of hibernating leafhoppers have demonstrated the same local differences in the species composition of populations shown by host collections, but usually with a larger number of species per hibernating sample than found in a host sample. On sycamore trees separated from others by a matter of miles, some leafhopper species may be absent and the species may be some time colonizing or re-colonizing the locality. This may be the explanation for the absence of *arta* and *usitata* from the isolated trees furnishing the Camargo collections (table 2), since both species of *Erythroneura* occur abundantly 10 miles away. As can be seen from figures 2 to 7, however, populations are sufficiently scattered through the complete range of each species that every suitable locality should have a potential supply of its possible sycamore components. This consideration and the fact that these leafhoppers swarm freely when entering or leaving hibernation lead me to believe that in this case vagility is a minor factor in affecting the distribution and abundance of these animals.

Ecological Adjustment

Weather

In looking over the sycamore collection records, the size of leafhopper populations appears to be correlated directly with humidity or rainfall. Exposed trees, or trees in a local drought area, usually have small populations, whereas trees in humid valleys almost invariably have large populations. It was mentioned above that large populations usually have the greatest diversity of species. From this we may conclude that in general high humidity favors the increase of *all* members of the *lawsoni* group. Apparently what is good for one species is also good for the others. The distribution maps of *usitata, torella, morgani,* and especially *bella* indicate that low temperature is one of the factors determining the northern limits of range of these species. In the case of *arta* and *lawsoni,* which extend northward beyond the limits of the other four species, higher average temperatures do not seem to have any deleterious effect in the more southern part of the range.

Temperature. An analysis of weather data compiled for Illinois (Page, 1949) shows that temperature and rainfall behave quite differently. Temperature is the more stable element of the two, figure 8. There is an average temperature difference of about 14° F. in winter and about 6° F. in summer between northern and southern Illinois, forming a moder-

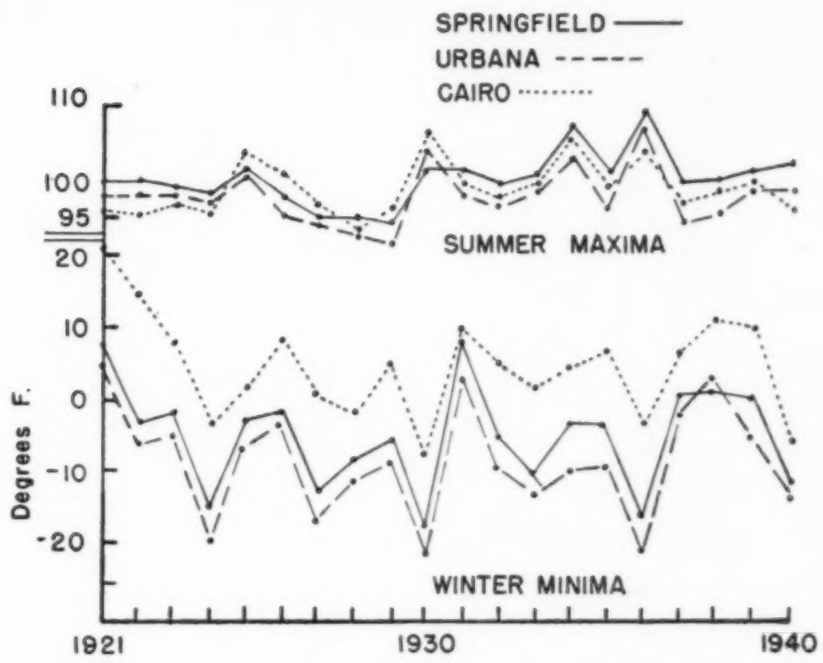

FIG. 8. Maximum (above) and minimum temperatures (below) for three Illinois localities for 1921–1940.

ately regular gradient from north to south. The winter extremes of northern and southern Illinois are about 15° apart, but the summer extremes are actually a little lower in southern Illinois, in spite of its higher summer average. There is thus no reason to consider that the more northern *E. arta* and *lawsoni,* would be at any disadvantage regarding high temperatures in the southern portion of their ranges.

The most important characteristic of Illinois temperatures is that, although they vary from year to year, they vary almost uniformly over the entire state. In a hot year temperatures are uniformly high; in a cool year they are uniformly low. Thus annual variation of temperature extremes alone could account for year to year fluctuations in leafhopper populations over the state as a whole but could in no way account for the irregular, local differences which are expressed on the distribution maps.

Precipitation. Precipitation has both direct and indirect effects on the leafhoppers. Directly, driving rains dislodge and destroy some nymphs, and possibly adults as well. Indirectly, however, precipitation is of much greater importance. It affects not only the succulence of the leaves on which the hoppers feed, but also the evaporation and transpiration of the foliage, which in turn affects the humidity inside the confines of the tree as a whole. Precipitation also affects the humidity of the general region, thus exerting a profound influence on the drying potential of winds.

Over Illinois as a whole the annual precipitation varies from about 32 inches in the north to 46 inches in the south. This differential is mainly the result of differences in winter precipitation. During the growing season (May through September) the precipitation is moderately even over the whole state. July and August are probably the most critical months for *Erythroneura* development, and in these two months the average Illinois precipitation is very nearly uniform over the entire state.

Precipitation, like temperature, varies from year to year, figure 9, but, unlike temperature, it varies to a remarkable degree from place to place. This is illustrated graphically by the 20 year record for July rainfall for Springfield and Urbana, Illinois, figure 10, and by the tran-

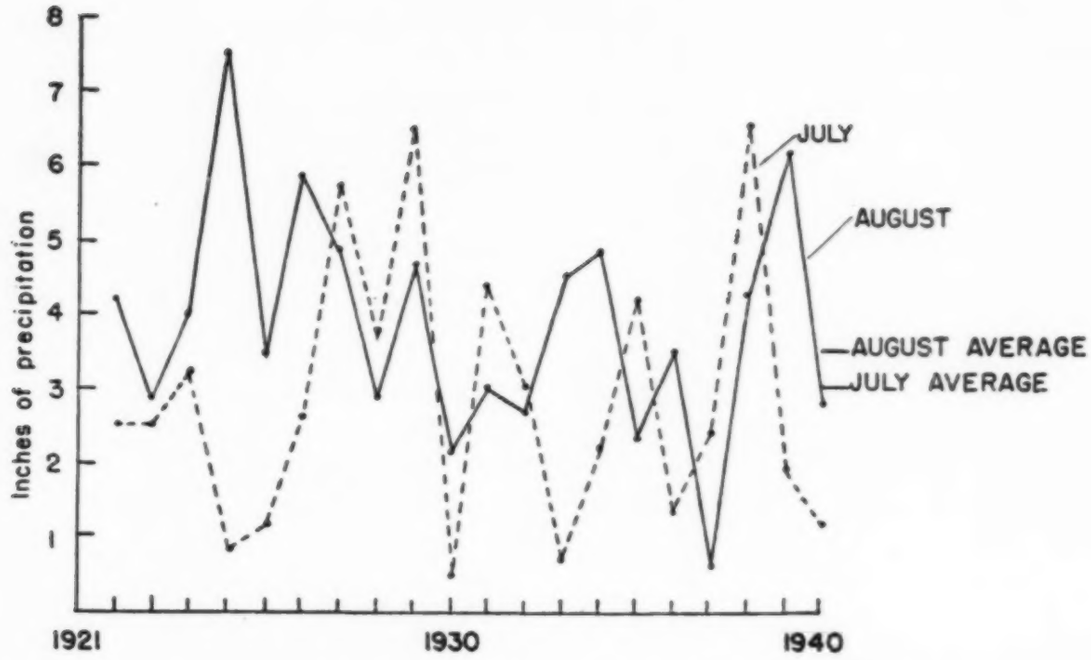

FIG. 9. July and August precipitation for Urbana, Illinois, 1921–1940. The long term average precipitation is indicated by the bars at the right.

sect of July and August rainfall for the same year across central Illinois, figure 11. In localities only a few miles apart, the rainfall may be well above average in one, below normal in the other, in the same season. Even in generally dry years the same local variation occurs. In the Illinois drought years of 1935–1938, for instance, August precipitation for the Grafton area (average 3.60 in.) was only .25, .94, .79, and 1.11 in., but for the Charleston area (average 3.52 in.) was 2.43, 2.87, 2.21, and 3.73.

Correlation with Leafhoppers. If we visualize the geographically undulating precipitation pattern illustrated in figure 11 projected in time in the variable fashion shown in figure 9, the result is a complex and irregular mosaic of hit-and-miss variation. This condition, varying locally

on this issue. C. D. M. would prefer a more or less subjective [illegible] ing information gained by the statistical method but modifying it with knowledge of probable actual phylogenies obtained in a variety of ways. He considers the [illegible] in inverse ratio to the size of the unit. The latter qualification will prevent the recognition of unjustified monotypic genera." Thus while *Proteriades* (species 1–24) was labeled a genus by C. D. M.,

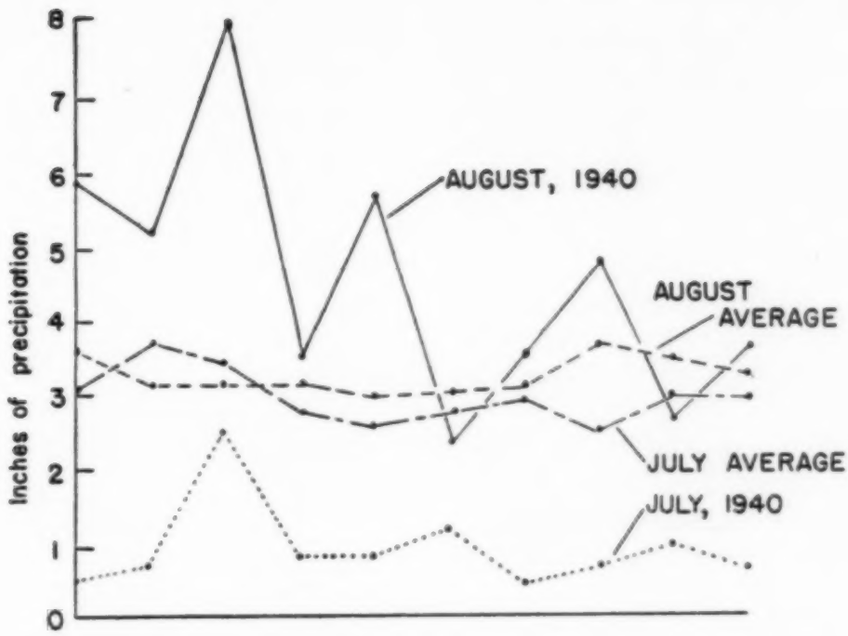

FIG. 11. Precipitation in a transect across central Illinois in July and August, 1940, compared with 40 year averages for each locality. The ten stations run from Quincy (at left) to Danville (at right).

from year to year and at any one locality more or less independently of another, is the same that we find in the distribution of species proportions in the sycamore leafhopper collections.

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both. He maintains that in the example cited above the nomenclature reflects descent in one case (by including *Dasyosmia* in the genus *Hoplitis*) but indicates divergence in another case (calling *Pro-*

ditions in which it is more successful than the others. All these conditions occur within the changing variability pattern of the weather over the sycamore range as a whole.

PARASITISM AND HOST PHYSIOLOGY

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for all four figures)

having them cut the ordinate at a point which would result in the number of categories (i.e., genera and subgenera) closest to the number established by

TABLE 2. *Collections of* Erythroneura *on identical sycamore trees in successive years.* The first collection number refers to rank of the species in the population, the second number to the actual percentage of the population represented by each species. Differences between the two collections are indicated in the third line for each locality.

		lawsoni	*arta*	*usitata*	*torella*	*morgani*	*bella*
Camargo	1954	1–73	0	0	2–26	3–1	0
	1955	1–56	0	0	2–41	3–3	0
		0–17			0–15	0–2	
E. Peoria	1953	1–65	3–11	5–2	2–13	4–9	0
	1955	1–53	3–13	2–18	5–4	4–12	0
		0–12	0–2	3–16	3–9	0–3	
Hennepin	1953	2–27	1–48	3–21	4–3	5–1	0
	1955	1–41	3–11	2–37	0	4–11	0
		1–14	2–37	1–16	1–3	1–10	
Mounds	1950	2–23	1–65	3–6	0	3–6	
	1953	1–34	2–25	3–19	4–16	5–6	0
		1–11	1–40	0–13	1–16	2–0	
Murphysboro	1953	1–31	2–29	0	4–8	3–24	4–8
	1955	2–19	1–56	3–9	0	4–8	4–8
		1–12	1–27	3–9	1–8	1–16	0–0
Newton	1953	1–51	2–22	4–8	5–5	3–13	0
	1955	1–50	2–17	3–14	5–6	4–13	0
		0–1	0–5	1–6	0–1	1–0	

decreases, until chiefly *lawsoni* occurs at the extreme edge of the group's tolerance for these factors.

CHANGE WITH TIME

Collections were made on the same trees in two different years to obtain some measure of the changes occurring in population proportions with time. In six of these sets, ecological conditions appeared similar at the time of both collections. A perusal of these six sets of data, listed in table 2, indicates that the same type of population differences occur from year to year on the same tree as were noted between different trees in the same vicinity at the same time (see table 1). In very few cases are the percentages between paired collections identical for any species.

In the Camargo and Newton sets, there was little or no change in standings, but the percentage of *lawsoni* dropped appreciably in 1955 compared to 1954. The two East Peoria collections demonstrate a reversal of position between *usitata* and *torella,* which is surprising in view of the relative stability of the rest of the population. In the Hennepin, Mounds, and Murphysboro collections the relative positions of four or five species changed. At Hennepin and Murphysboro, the position of *lawsoni* shifted from second to first, and from first to second, respectively. In the Murphysboro collection a simple first-second switch occurred between *lawsoni* and *arta,* whereas in the Hennepin collection *arta* dropped from first in 1953 to a poor third behind *lawsoni* and *usitata.*

These collections indicate the occurrence of population fluctuations from year to year which are variable in different localities both as to degree of change and the species involved.

Applying these results to the maps in figures 2 to 7, it is obvious that there is represented here only a cross-section of a highly plastic series of populations. It follows that maps based on data collected in other years would differ from the present ones in details concerning a given locality. It is probable, however, that taken as a whole these data would give the same over-all proportions as shown in figure 1.

Explanation of Results

The foregoing analysis of these sycamore populations demonstrates that the proportions of the various species differ in an apparently haphazard fashion (1) from place to place and (2) from year to year. In an attempt to explain such variability, three factors would normally be supposed to be operating: interspecific competition, vagility (ability to disperse), and ecological adjustment.

Competition

The data accumulated on the six species of the *lawsoni* group indicate that interspecific competition was not a factor affecting either their abundance or distribution during the period when these collections were made. Due to differences in collecting efficiency depending on wind velocity and temperature, and the difficulty in obtaining large samples from tall trees or those crowded in with other tree species, it is virtually impossible to devise a good method for determining the absolute abundance of leafhoppers per unit tree or unit area. It is easy, however, to estimate populations as small (roughly 1 to 50 leafhoppers obtained per medium-sized tree), medium (roughly 50 to 500 per tree), and large (over 500 per tree, often reaching many thousands); these are the categories used here. In this section I refer also only to the southern half of Illinois, where all six species are found as a coexisting complex.

If interspecific competition were a factor influencing the relative abundance of the sycamore leafhoppers, we would expect trees with small populations, where competition would be at a minimum, to have the greatest average diversity of leafhopper species. Conversely we would expect trees with the largest leafhopper populations, where competition would be greatest, to have the least diversity of species. But this is not what we find. Regardless of size or situation, trees with small populations usually harbor only *lawsoni* or *arta*, whereas trees with large populations almost always support all six species. I am therefore led to believe that interspecific competition is not a factor in regulating populations of these species.

This same phenomenon is illustrated strikingly by some other groups of *Erythroneura*. The shingle oak (*Quercus imbricaria*) has a large and restricted fauna of about a dozen species of *Erythroneura*. In most localities we have visited, it is difficult to obtain more than a dozen or two specimens from a clump of shingle oaks. Each collection usually has only two or three species. Certainly there is no competition between leafhoppers here, because a single small leafhopper may have a whole tree to itself. On one occasion we encountered a shingle oak which was "loaded," having many hundreds of leafhoppers; this collection contained every species ever recorded from the host. Similar examples could be cited for several other hosts on which we have collection records.

What appears to be interspecific competition does occur occasionally between these leafhoppers and the sycamore tingid or lacebug, *Corythuca ciliata* Say, which is also widespread on sycamores in Illinois. In 1955 in certain Illinois localities (Oregon and Kankakee), the tingids reached such a high population level that the sycamore leaves became coppery and brittle. *Erythroneura* were either absent or very scarce on such trees, many of

them so situated that they should have had a good *Erythroneura* fauna. It is likely that the excessive tingid feeding simply dried the leaves to such an extent that the *Erythroneura* could not get sufficient food out of them, and either left the host tree or died. We have too few data to know if this tingid action has a differential effect on different species of *Erythroneura.*

In contrast to the sycamore collections reported here, instances are known in which *Erythroneura* have completely etiolated their host. I have seen redbud trees (*Cercis canadensis*) in this condition and also on one occasion in Columbus, Ohio, a series of etiolated sycamore trees. These latter sustained immense numbers of *Erythroneura lawsoni.* Cultivated Virginia creeper and grape may be so severely etiolated by various species of *Erythroneura* that the leaves wither and fall in midseason. Under these conditions some sort of competitive effect or differential species survival may occur. That such conditions occur very rarely on sycamore, however, is shown by the fact that in ten years of collecting I have personally encountered only the single example of etiolation cited.

It seems certain, therefore, that if some form of interspecific competition occurs between the sycamore leafhoppers, it is extremely local and/or infrequent.

Vagility

The effect of vagility on *Erythroneura* populations is obscure. Collections of hibernating leafhoppers have demonstrated the same local differences in the species composition of populations shown by host collections, but usually with a larger number of species per hibernating sample than found in a host sample. On sycamore trees separated from others by a matter of miles, some leafhopper species may be absent and the species may be some time colonizing or re-colonizing the locality. This may be the explanation for the absence of *arta* and *usitata* from the isolated trees furnishing the Camargo collections (table 2), since both species of *Erythroneura* occur abundantly 10 miles away. As can be seen from figures 2 to 7, however, populations are sufficiently scattered through the complete range of each species that every suitable locality should have a potential supply of its possible sycamore components. This consideration and the fact that these leafhoppers swarm freely when entering or leaving hibernation lead me to believe that in this case vagility is a minor factor in affecting the distribution and abundance of these animals.

Ecological Adjustment

Weather

In looking over the sycamore collection records, the size of leafhopper populations appears to be correlated directly with humidity or rainfall. Exposed trees, or trees in a local drought area, usually have small populations, whereas trees in humid valleys almost invariably have large populations. It was mentioned above that large populations usually have the greatest diversity of species. From this we may conclude that in general high humidity favors the increase of *all* members of the *lawsoni* group. Apparently what is good for one species is also good for the others. The distribution maps of *usitata, torella, morgani,* and especially *bella* indicate that low temperature is one of the factors determining the northern limits of range of these species. In the case of *arta* and *lawsoni,* which extend northward beyond the limits of the other four species, higher average temperatures do not seem to have any deleterious effect in the more southern part of the range.

Temperature. An analysis of weather data compiled for Illinois (Page, 1949) shows that temperature and rainfall behave quite differently. Temperature is the more stable element of the two, figure 8. There is an average temperature difference of about 14° F. in winter and about 6° F. in summer between northern and southern Illinois, forming a moder-

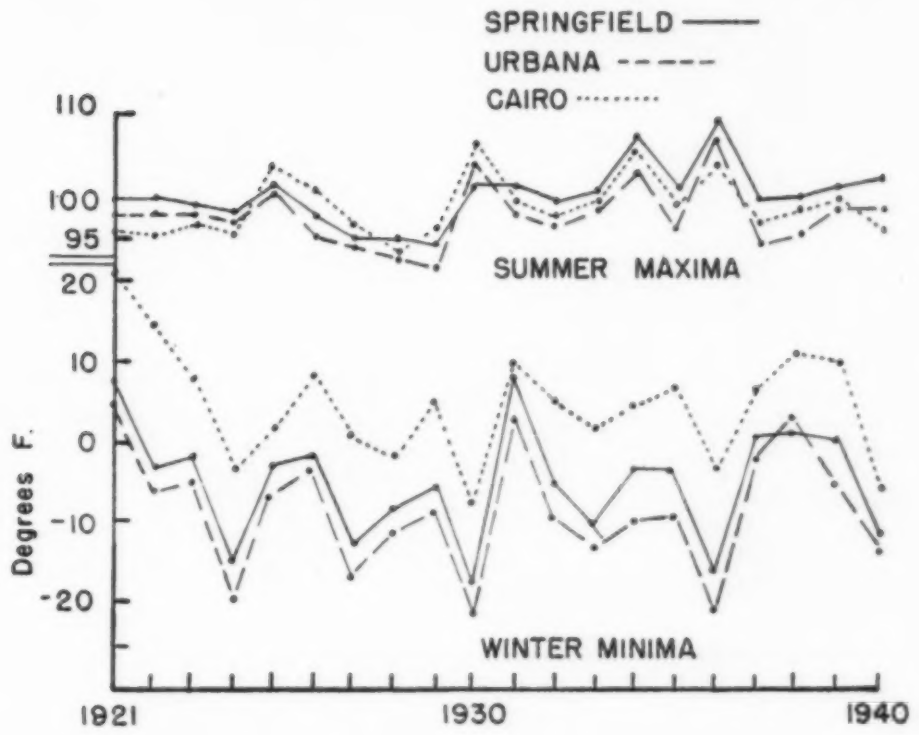

FIG. 8. Maximum (above) and minimum temperatures (below) for three Illinois localities for 1921–1940.

ately regular gradient from north to south. The winter extremes of northern and southern Illinois are about 15° apart, but the summer extremes are actually a little lower in southern Illinois, in spite of its higher summer average. There is thus no reason to consider that the more northern *E. arta* and *lawsoni,* would be at any disadvantage regarding high temperatures in the southern portion of their ranges.

The most important characteristic of Illinois temperatures is that, although they vary from year to year, they vary almost uniformly over the entire state. In a hot year temperatures are uniformly high; in a cool year they are uniformly low. Thus annual variation of temperature extremes alone could account for year to year fluctuations in leafhopper populations over the state as a whole but could in no way account for the irregular, local differences which are expressed on the distribution maps.

Precipitation. Precipitation has both direct and indirect effects on the leafhoppers. Directly, driving rains dislodge and destroy some nymphs, and possibly adults as well. Indirectly, however, precipitation is of much greater importance. It affects not only the succulence of the leaves on which the hoppers feed, but also the evaporation and transpiration of the foliage, which in turn affects the humidity inside the confines of the tree as a whole. Precipitation also affects the humidity of the general region, thus exerting a profound influence on the drying potential of winds.

Over Illinois as a whole the annual precipitation varies from about 32 inches in the north to 46 inches in the south. This differential is mainly the result of differences in winter precipitation. During the growing season (May through September) the precipitation is moderately even over the whole state. July and August are probably the most critical months for *Erythroneura* development, and in these two months the average Illinois precipitation is very nearly uniform over the entire state.

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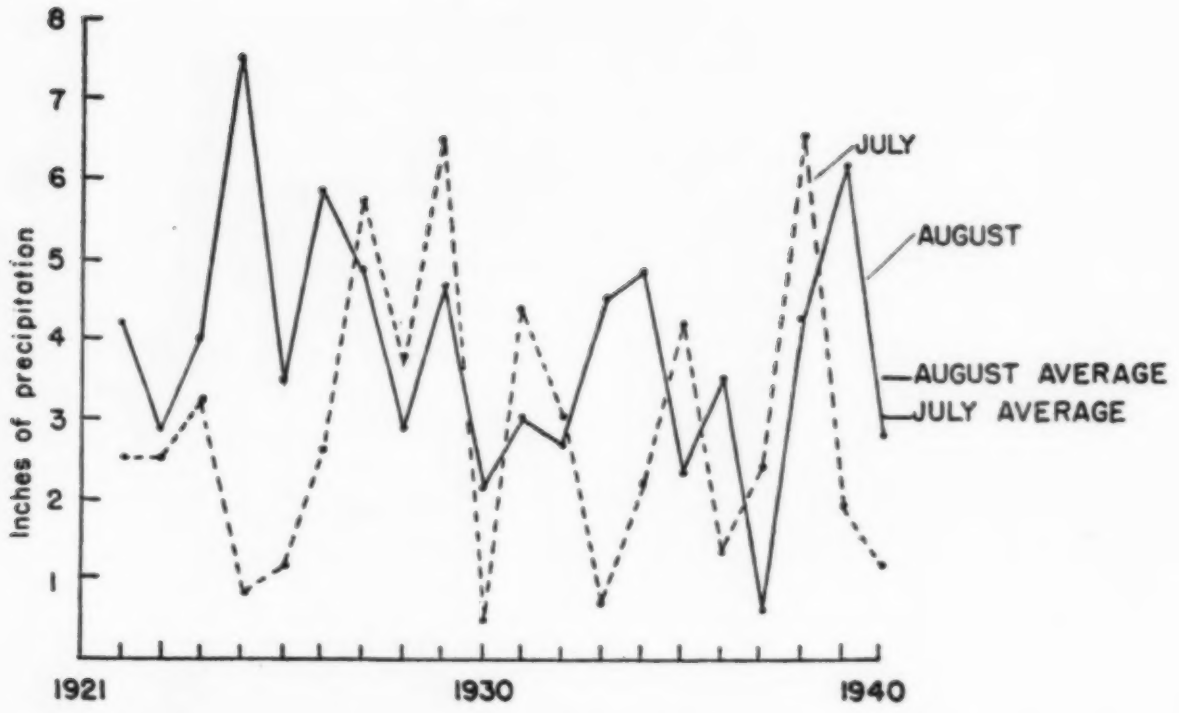

FIG. 9. July and August precipitation for Urbana, Illinois, 1921–1940. The long term average precipitation is indicated by the bars at the right.

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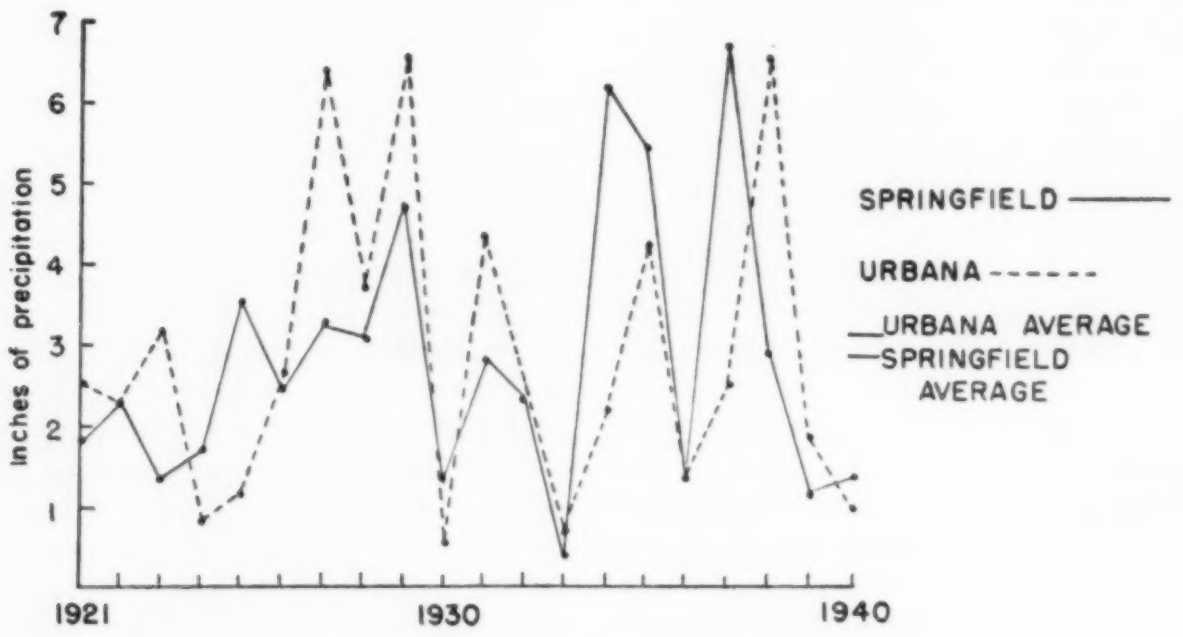

FIG. 10. July precipitation at Springfield and Urbana, Illinois, 1921–1940. The long term average July precipitation for each locality is shown by the bars at the right.

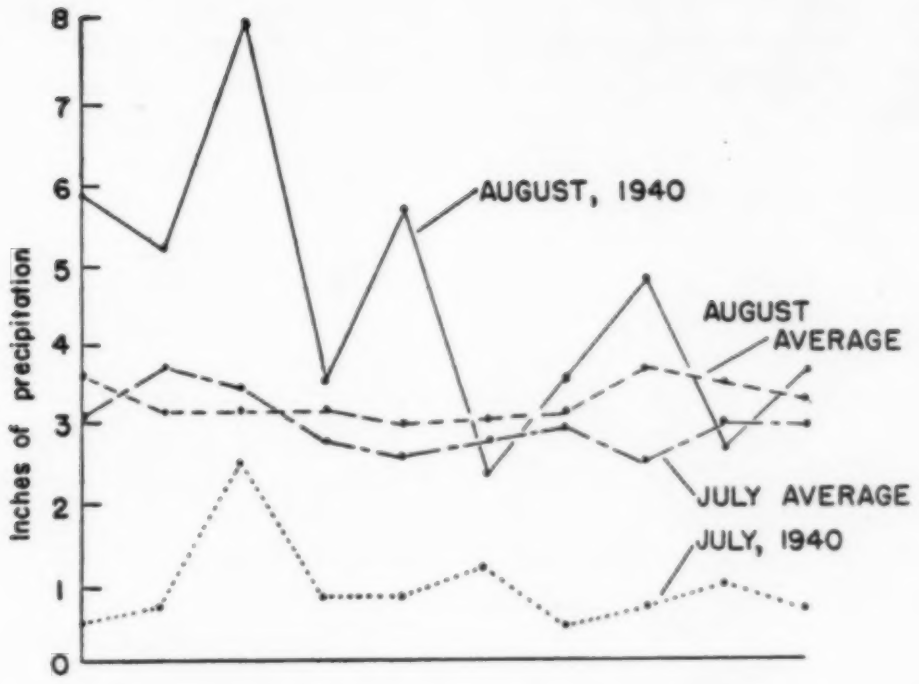

FIG. 11. Precipitation in a transect across central Illinois in July and August, 1940, compared with 40 year averages for each locality. The ten stations run from Quincy (at left) to Danville (at right).

from year to year and at any one locality more or less independently of another, is the same that we find in the distribution of species proportions in the sycamore leafhopper collections.

Annual variations in temperature superimposed on these conditions add another dimension to the weather variation. Unfavorable extremes have a drastic effect on the leafhoppers. Following excessive hot, dry spells we have repeatedly found *Erythroneura* nymphs dead on the leaves of their hosts, apparently anchored by their beaks. After searing winds *Erythroneura* are difficult to find except in narrow, damp ravines. During 1955, only small populations of these insects were obtained from sycamores in the parts of central Illinois where corn yields were materially reduced by local drought conditions.

The leafhopper records indicate that with the areas of unfavorable conditions are intermingled areas of favorable ones which have resulted in local increases in leafhopper populations. Since five of the species are predominant in some collections, each of these species must encounter its own peculiar optimum or at least conditions in which it is more successful than the others. All these conditions occur within the changing variability pattern of the weather over the sycamore range as a whole.

PARASITISM AND HOST PHYSIOLOGY

Species of *Erythroneura* are frequently parasitized by hymenopterous wasps of the family Dryinidae. Such parasitism has been noticed among leafhoppers in sycamore samples but neither its intensity nor species selectivity was recorded. The parasitism was sporadic and appeared to be random, hence of little importance in this study. It is possible, however, that the parasitism is selective, at least under certain conditions. This has been found true with some other insects (Allee, Emerson, Park, Park, and Schmidt, 1949). Detailed studies of this subject might prove of great interest concerning this *Erythroneura* problem.

Although physiological differences between individual host trees might constitute a factor having some effect on the populations of species of *Erythroneura* occurring on any one tree, there appears to be no clear-cut evidence that such is

the case in these studies. To the contrary the duplicate collections summarized in table 2 indicate that species may come and go on the same tree in different years. There is, of course, always the possibility that detailed observations designed to investigate this point may uncover interesting and unforeseen results.

If parasitism and host physiology should prove to be selective factors in determining relative abundance of the sycamore leafhoppers, they would undoubtedly be smaller scale variables added to the major variables of weather. Furthermore, both parasitism and host reaction would themselves come under the influence of weather variation.

Concept of a Niche

From the data presented in the preceding pages it is clear that even as restricted a niche as the leaves of a single species of tree, considered over its entire range, is a complex, variable unit. Because of annual fluctuations in temperature combined with both local and annual fluctuations in precipitation, modified locally by differences in terrain, the range is a kaleidoscope of changing ecological conditions. To these changes are added other complicating and presumably equally varying factors such as parasitism and host physiology. Random parts of the range become unfavorable at the same time that other parts remain or become favorable. The periphery of the range would represent a statistical or chance function of time and the distance between favorable areas integrated with the vagility of the species.

It is probable but not necessary that some parts or areas of the range are permanently favorable to the species. In times of severest ecological inimicality, these areas would be the primary dispersal points for the recolonization of denuded areas when these latter returned to a condition favorable to the species. It is also probable that as a rule over much of the range, ecological conditions vary within the acceptable range for the species, resulting in fluctuations in population size rather than in extirpation.

The narrower the ecological tolerance or the vagility of the species, the more it would tend to become restricted to those areas which were permanently favorable to it. The wider the ecological tolerance and the greater the vagility, the more the species would colonize peripheral or outlying areas during seasons of widespread favorable conditions.

In these concepts of weather and range I believe we have all the elements necessary to explain the distribution of the sycamore leafhoppers. Interspecific competition within the genus does not seem to be an important factor with these insects, hence each species increases or decreases in number independently of population changes in the other species. With the rarer species, peripheral areas may at times be highly favorable on a local basis for population increase, resulting in the pecularities of range noted especially in the case of *usitata* and *torella*. Each species has a slightly different ecological optimum and hence a variety of rates of population increase under the various conditions found in different localities. As a result, the rankings of the various leafhopper species in the sycamore population are a function of the ecological history of the locality, modified (through vagility values) by the history of adjacent localities.

Principles of Coexistence

Earlier in this paper I mentioned that numerous groups of insects seem to illustrate the phenomenon of the coexistence of several species. So closely do large numbers of these groups parallel conditions found in the sycamore leafhoppers, that such coexistence would seem to be the rule for these small organisms. From the observations on the sycamore leafhoppers, it is possible to deduce the following generalizations regarding coexistence:

1. An ecological niche or habitat which serves as the range of a species has a

dimension in both time and space. Normally such a niche is not ecologically uniform, but is a changing kaleidoscope of local and annual variations around certain ecological means. The niche must be large enough to allow the species to maintain a viable reproducing population during periods of ecological pessimum.

2. Such a niche may be occupied and used by more than one species at the same time. If so, the size of each occupant species population will independently tend to increase or decrease in various areas of the niche depending on pertinent ecological changes locally and annually. I would not consider as coexistence cases in which there was a seasonal succession in the use of the niche by different species.

3. Because no two species are exactly alike genetically and therefore physiologically, each species should theoretically have an ecological optimum slightly different from all other species. It follows that, in cases of multiple occupancy of a niche, each species will tend to become abundant at a time or place different from the other species. As a corollary, at any given time, as one species would approach its maximum population size in certain areas of the niche, other species would do so in other areas. These areas might shift with time.

4. When interspecific competition for food or space is not the chief factor governing density of populations of coexisting species, there appears to be no theoretical limit to the number of species which can occupy the same niche. In insect groups we have records of many cases in which four to a dozen or more species inhabit a single niche. The survival of any one species would depend solely on its ability to maintain viable populations, rather than on its ability to become dominant in the population. A good example is *Erythroneura bella,* figure 7.

5. If interspecific competition for food or space were the chief factor governing density of populations, then theoretically the prerequisite for the survival of any one of the coexisting species would be its ability to develop larger populations than the others of its companion species in some portion of the ecological variability pattern of the niche. The coexisting complex would therefore be composed of species each with a different optimum and each outcompeting all the others under conditions of its own optimum. If interspecific competition occurred consistently between the species of sycamore leafhoppers (compare figures 4 to 9), only *lawsoni, arta, usitata, torella,* and possibly *morgani* would have a chance of surviving; *bella* would theoretically become extinct.

Considering the food problem of maintaining large enough populations to preserve reproducible units, the number of species which could occupy the same niche would be inversely proportional to the food requirements, hence usually to the absolute size, of the organism. In these insect examples I have cited, the individual food requirement is small. It is difficult to visualize half a dozen species of elephants all maintaining reproducible units if restricted to sycamores for food. On the other hand, the tropical forests described by Dobzhansky (1950), containing hundreds of species of trees growing in the same small plot of ground, would seem to be outstanding examples of competitive natural coexistence involving organisms of large size.

Adaptive Radiation

Even if it acted as a selection pressure at only irregular intervals, interspecific competition would theoretically exert selection pressures which would make each joint occupant of a niche more and more narrowly adapted to the ecological vicinity of its optimum within the niche. The same selection pressures would tend to move the optima of the various species farther apart. Such a restriction would lead, in the terms of Simpson (1953), to a subdivision of the zone (niche). It is my belief that such a narrowing of ecological tolerances accompanying inter-

specific competition is the usual forerunner of adaptive radiation. In other words, after a group of coexisting species become specialized for different ecological combinations of the niche, each species may then evolve in new directions because of different selection pressures imposed by the more restricted nature of its environment.

Interarea Mixing

In times when geological or climatic changes allow the dispersal of species from one area into another, there may occur a considerable increase in the number of species occurring in the same niche. Simpson (1953) has outlined the course of competition and extinction which occurs between competing members of such groups. An entirely different condition would prevail in the case of coexisting species between which no interspecific competition occurred. In the case of these species, intercontinental dispersal would simply increase the number of species which would share the same niche.

Simpson's examples are drawn primarily from the Mammalia, which have a relatively large size and consequently large individual demands on the environment. With organisms such as the greater portion of the insects, their small size enables even a small niche to sustain a moderately large number of species. If the smaller organisms were actually competing they would of course follow in general the outlines of Simpson's proposed interplay, but a larger relative number of species probably would finally survive than in the case of larger organisms.

Viewed in this light, interarea mixing of biotas is a powerful agent in increasing the instances and complexity of natural coexistence, and thus in setting the stage for additional possible cases of adaptive radiation.

Summary

1. Studies made in Illinois of the sycamore leafhoppers, a monophyletic group of six species comprising the *lawsoni* complex of the genus *Erythroneura,* show that several species may occupy the same ecological niche at the same time.

2. The relative abundance of each species varies in haphazard fashion both locally and annually over the entire range of the species, with high points occurring even along the periphery of the range. In five of the six species, each species occurs as the dominant one in some of the collections made.

3. There is evidence that interspecific competition did not occur between these organisms during the period of this study. General observations on the group indicate that in the entire genus *Erythroneura* interspecific competition, if it occurs at all, is sporadic and local.

4. The most important ecological factors affecting these insects appear to be rainfall and shelter, or more precisely, humidity and host succulence. Rainfall appears to be the most important factor. It varies from place to place and from year to year in the same haphazard pattern as do the sycamore leafhopper records, and is suggested as the basic reason for the peculiarities in the relative abundance of the leafhoppers.

5. On the basis of these observations the following general principles are advanced:

a. An ecological niche or habitat serving as the range of a species is normally not uniform, but is a changing kaleidoscope of local and annual variations around certain ecological means.

b. Species are always at least slightly different physiologically, and any two will therefore have slightly different ecological optima.

c. As a result of principles *a* and *b,* such a niche may be occupied and used by more than one species at the same time.

d. In the absence of interspecific competition, there appears to be no predictable limit to the number of species which can occupy the same niche.

e. Under conditions of interspecific competition, a species would survive only

if, under conditions of its own optimum, it could develop larger populations than its companion species.

6. Interspecific competition, when it occurs, probably causes the optima of coexisting species to move farther apart and lead to a condition conducive to adaptive radiation.

7. Interarea mixing of biotas increases the instances and complexity of natural coexistence, and in this way sets the stage for additional possibilities of adaptive radiation.

Acknowledgments

Many of the ecological ideas implicit in this paper run parallel to points emphasized by Andrewartha and Birch (1954), Allee, Emerson, Park, Park, and Schmidt (1949), and Allee and Schmidt (1951), to name only a few. Many of the ideas in this paper also have their foundations in works of many writers on evolution, notably Simpson and Wright. I wish to express my appreciation to the tremendous help received from these and other sources in the literature.

I am also indebted for a great deal of help, both in collecting material and in the discussion of various points, to members of the staff of the Illinois Natural History Survey, especially Doctors H. B. Mills, T. E. Moore, M. W. Sanderson, R. B. Selander, P. W. Smith, and L. J. Stannard.

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A QUANTITATIVE APPROACH TO A PROBLEM IN CLASSIFICATION [1]

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INTRODUCTION

The purpose of the study reported in this paper was to determine whether certain statistical procedures might aid persons interested in the relationships among organisms. The objectives of our study were to investigate numerous characters simultaneously in a considerable group of species; to quantify the relations shown among the species, using objective methods; and to indicate these relationships. The organisms selected as an example for use in this study are solitary bees in the family Megachilidae. This choice was made because one of us (C. D. M.) has made recent systematic studies of these insects, so that conclusions as to the relationships obtained by usual systematic procedures could be compared with the results of the methods described below.

A major concern of the systematist, when he devises a classification, is to make one in accordance with probable phylogeny, but first he must assess the characters and determine relationships in a static or nonhistorical sense. Later he may decide on the most likely lines of descent. The greater part of this paper concerns the first step, determination of static relationships, although ideas as to phylogeny are, of course, included.

In the absence of data from fossils, and sometimes even when these are available, systematists often differ widely as to relationships among organisms. This is true even if problems raised by convergence and varying rates of evolution can be settled or temporarily ignored. It is easier to appreciate differences than similarities. Therefore, in most groups, the species are differentiated long before serious studies of relationships begin. A systematist who is interested in phylogeny, but lacks paleontological data, utilizes character correlations observed in the course of his study in determining relationships and delimiting the levels (genera, families, etc.) in the hierarchy of classification.

Weighting of Characters

Each species has an enormous number of characters. The systematist normally tries to use as many as he can find, but is hampered in his study of character correlations by the limitations of the human mind. Also, he is necessarily subjective in his choice and weighting of characters.

If he notes that certain characters vary erratically with respect to one another

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The ideas presented herein were first tried out on informal groups, the "Biosystematists" and "Evolutionists," at the University of Kansas; we received worthwhile suggestions from various members of each group.

We wish also to acknowledge contributions from the University of Kansas General Research Fund for assistance.

among various species, the systematist will regard the numerous combinations of such characters as valuable in differentiating species, but will not use these characters in erecting higher categories, such as genera. On the other hand, when he finds several characters, which are not related by function or other obvious causal mechanisms, consistently associated with one another among several species, and a quite different combination of characters associated in another group of species, he feels that he has found evidence of two species clusters and probably of two phyletic lines. The more numerous are the differentiating characters associated within each group, the more confident is he of the conclusion. Even though they may be minute and inconspicuous, the systematist will weight such characters heavily in comparison with those which vary from species to species with no obvious relation to one another. His judgments as to relationships, and hence higher categories, will therefore be largely based upon a few characters which he finds to be perfectly associated. Especially if the number of such characters is small, the discovery of additional character associations may alter judgment about relationships. Sometimes, as a result of parallel evolution, two or more sets of associated characters may suggest different relationships. In this case the systematist must decide, on the basis of experienced judgment or mere intuition, which set of characters indicates phyletic relationships. In brief, he will give greater weight to the characters that he believes most significant.

The problem of weighting characters is important, but it is not obvious how the weighting should be done. No doubt the so-called conservative characters should be heavily weighted, but how are they to be recognized with certainty? A systematist can often give sound reasons for his judgments on this matter; as indicated above, they usually have to do with character associations. On the other hand, his judgments are often quite subjective so that the development of a classification may be more of an art than a science.

Weighting might be according to the number of genes influencing a character, but no one knows how many genes are involved in the formation of most characters of even the genetically best known organisms; therefore, this method of weighting is useless. Even if we knew the number of genes affecting a character, this would not give us a basis for evaluating its importance in judging relationships.

It is often said that "purely adaptive" characters are unlikely to be conservative and hence should receive less weight in studies of relationships than characters having no obvious adaptive value. Thus, among a group of species, several may have independently invaded a particular habitat and independently evolved the same adaptive character. This situation becomes obvious if conservative characters are found which show the true phylogeny. On the other hand, it is often evident that obviously adaptive features do characterize large groups of species and hence do indicate phyletic relationships. It is also probable that every character, if not itself adaptive, is at least determined by genes which have adaptive value through some other influence. For these reasons we cannot use adaptiveness, even if there were a way to measure it, as a criterion for weighting.

We are left with the conclusion that, in the absence of fossils, constant association of several characters which are not related causally is the best evidence of the conservatism of these characters. If only a few characters are used, studies of relationships require weighting of certain conservative characters in the mental processes of the systematist. By using a large number of characters and species, however, we believe that weighting becomes unnecessary because the magnitude of correlation coefficients calculated between species would be little affected except by extreme weighting.

Kinds of Characters

Almost all of the characters used in our study are morphological or color features of the adult insects. One of us (Michener, 1953) has emphasized the value of characters of other stages (e.g., larvae, pupae), of behavioral characters, etc., but also has stressed that these are merely additional characters, of neither more nor less importance than other adult morphological characters that might be found. We have used external adult characters because they are the only ones known for most of the species concerned. Also, since a very large number of them were considered, we believe that the use of additional characters might not have materially altered our results, because additional characters have a decreasing effect on the index of relationship which we have used.

We believe that our morphological characters represent, in a genetic sense, a random sample of all the characters, and thus a random sample of genetic variation. There are no special groups of morphological genes. Many genes influence morphological as well as behavioral or physiological characters.

Quantification of Relationships

The methods described below make possible a certain degree of quantification of similarities, using many more characters than are ordinarily employed in a systematic study. This means that we can give some measure of the amount of evolutionary change that has occurred since phyletic lines diverged. It is our basic assumption that degree of difference can give an estimate of evolutionary divergence (E). E is clearly the product of $r \times t$, where r is the rate of evolution (dE/dt) and t is time. From an estimate of E, there is no way to obtain r or t; it is, for example, quite probable that one pair of species diverged quite recently compared to another equally different pair.

Possible parallelisms present a difficult problem. The amount of evolution, after divergence from a common stem, of two forms that have evolved in a parallel manner, would be underestimated by our method, as well as by customary systematic methods. In the majority of cases in animals, however, parallelisms probably involve only a few characters. Thus in a study of parallelisms in saturniid moths (Michener, 1949a) many structures were found to appear or disappear independently in various genera but in no instance did a large group of associated characters behave in this fashion. Among the bees used in the present study there is evidence of acquisition of clear wings, red integumental pigment and abundant pale pubescence by various unrelated species in desert areas. These tendencies could affect a maximum of only five of the 122 characters studied. We believe that much of the difficulty due to parallelisms is eliminated in our study by the large number of characters used. Parallelism or convergence affecting only a few characters would not greatly influence our measure of relationship.

It is impossible to show all details of relationships by any hierarchy of systematic categories unless the latter are excessively numerous. Therefore, it is common for a systematist to state that a certain genus is closely related to another and that these two are more distantly related to others within a tribe or subfamily. He does not try to indicate all such details of relationships by the usual classificatory system. The systematist himself will be quite aware of this situation and may explain it, but to the uninitiated user of his work, the hierarchy may constitute something of a strait jacket for thoughts on relationships.

To indicate relationships more precisely than is possible by means of systematic categories, various diagrammatic schemes have been adopted. The commonest such scheme is a phylogenetic tree. Such trees are most justifiable if abundant and well distributed fossil evidence supports them. In the absence of such evidence we prefer to call such trees diagrams of relation-

ships. The dangers of such diagrams have been explained by many authors, but diagrams of relationships continue to be useful, and as yet no better method of indicating relationships has been devised for general use. We have therefore presented many of our results as diagrams of relationships. The details of the meanings of such diagrams will be discussed later.

The roles of the two authors may be described as follows: The basic data were gathered from previous publications and from specimens by C. D. M. After considerable discussion of methods, the statistical work (described in detail by Sokal and Michener, 1957) was carried out by R. R. S. and assistants without reference to C. D. M.'s systematic knowledge of the group. Thus, the relationships determined from statistical procedures could not have been biased by prior views of relationships based on ordinary systematic methods.

It may be suggested that bias has been introduced by our use of the same characters that were used in previous systematic studies. As other characters are not known, we were, of course, forced to use these. Moreover, it is our belief that any large group of characters would indicate similar relationships since, as we have said, we believe these characters to represent a random sample of the genetic variation.

Materials

The *Hoplitis* Complex and Its Older Classification

The bees used in this study are the American species of the megachilid genera *Ashmeadiella, Anthocopa, Hoplitis,* and *Proteriades.* These four genera, for convenience, may be called the *Hoplitis* complex; they are part of the genus *Osmia* as it is understood by European workers. A total of 97 species were included in the study after the few forms known from only one sex were eliminated. We do not believe that exclusion of such forms affects the usefulness of this work as a pilot study to determine the suitability of the method. There must have been many species which are now extinct, which can never be included in any such study. Therefore, the exclusion of certain other species should not affect our evaluation of the method. Fortunately, among the 97 included species, there is at least one representative of every subgenus or species group recognized in the earlier systematic studies.

As has been explained elsewhere (Michener, 1941; Hurd and Michener, 1955) the genera of the *Hoplitis* complex are closely related. Species of two of them (*Hoplitis* and *Anthocopa*) are found in Eurasia (with a few species in Africa), but with one exception Eurasian forms belong to different subgenera than American forms. We believed this complex particularly appropriate for our study because previous investigations indicated that it contains some large groups of very similar species as well as some apparently isolated species.

The bees of this group are solitary rather than social; each female is a reproductive, there being no worker caste. Nests are made in the soil or in wood or stems. Most of our species inhabit western America although several occur in the east; none is known south of Mexico.

Table 1 (the formal classification) shows the names and code numbers of the 97 species and the way in which they were organized into genera, subgenera, and species groups prior to the present study.

This classification is merely an attempt to place conveniently within the framework of a classificatory hierarchy, the ideas which are indicated with more accuracy and detail in the diagrams of relationships (figs. 1 and 2). These figures show relationships among the species groups including various details which cannot be indicated in the formal classification. Citations to the systematic papers which form the basis for these figures and table appear in Michener (1949b) and in Hurd and Michener (1955), with some

TABLE 1. *Formal classification of 97 species in the* Hoplitis *complex*

Genus group	Genus	Subgenus	Species group	Species code number and name
1.	*Proteriades*	*Cephalapis*	—	1. *jacintana* (Cockerell)
		Proteriades s. str.	—	2. *semirubra* (Cockerell) 3. *deserticola* Timberlake & Michener
		Xerosmia	A	4. *xerophila* (Cockerell)
			B	5. *nanula* Timberlake & Michener 6. *pygmaea* Timberlake & Michener 7. *reducta* Timberlake & Michener 8. *cryptanthae* Timberlake & Michener 9. *caudex* Timberlake & Michener 10. *hamulicornis* Timberlake & Michener 11. *truicauda* Timberlake & Michener 12. *bidenticauda* Timberlake & Michener 13. *nigrella* Michener 14. *tristis* Michener 15. *palmarum* (Cockerell) 16. *seminigra* Timberlake & Michener
			C	17. *remotula* (Cockerell) 18. *incanescens* (Cockerell)
	Hoplitis	*Hoplitina*	—	19. *howardi* (Cockerell) 20. *bunocephala* Michener 21. *bullifacies* Michener
		Acrosmia	—	22. *laevibullata* (Michener) 23. *rufina* Michener
		Andronicus	—	24. *cylindrica* (Cresson)
		Monumetha	A	25. *fulgida* (Cresson) 26. *louisae* (Cockerell) 27. *viridimicans* (Cockerell)
			B	28. *albifrons* (Kirby)
		Dasyosmia	—	29. *biscutellae* (Cockerell) 30. *paroselae* Michener
		Cyrtosmia		31. *hypocrita* (Cockerell)
		Alcidamea	A	32. *producta* (Cresson) 33. *pilosifrons* (Cresson) 34. *grinnelli* (Cockerell) 35. *colei* (Crawford) 36. *elongaticeps* Michener 37. *uvulalis* (Cockerell)
			B	38. *sambuci* Titus 39. *truncata* (Cresson)
		Formicapis	—	40. *clypeata* (Sladen)
		Robertsonella	—	41. *simplex* (Cresson)

TABLE 1—*Continued*

Genus group	Genus	Subgenus	Species group	Species code number and name
2.	*Anthocopa*	*Atoposmia*	A	42. *triodonta* (Cockerell) 43. *hebitis* Michener 44. *pycnognatha* Michener
			B	45. *oregona* Michener 46. *anthodyta* Michener 47. *panamintensis* Michener 48. *elongata* (Michener) 49. *abjecta* (Cresson)
		Hexosmia	—	50. *copelandica* (Cockerell)
		Eremosmia	A	51. *hemizoniae* (Cockerell) 52. *viguierae* (Cockerell) 53. *enceliae* (Cockerell)
			B	54. *rubrella* Michener 55. *hurdiana* Michener
			C	56. *daleae* Michener 57. *beameri* Michener 58. *mirifica* Michener 59. *hypostomalis* Michener 60. *nitidivitta* Michener 61. *rupestris* (Cockerell) 62. *robustula* (Cockerell)
	Ashmeadiella	*Chilosima*	—	63. *holtii* Cockerell 64. *rhodognatha* Cockerell
		Ashmeadiella s. str.	A	65. *altadenae* Michener 66. *rufitarsis* Michener
			B	67. *titusi* Michener 68. *femorata* (Michener) 69. *truncativentris* Michener
			C	70. *rufipes* Titus 71. *prosopidis* (Cockerell) 72. *sonora* Michener 73. *occipitalis* Michener 74. *bucconis* (Say) 75. *cactorum* Cockerell 76. *bigeloviae* (Cockerell) 77. *difugita* Michener 78. *leucozona* Cockerell 79. *foveata* Michener 80. *gillettei* Titus 81. *maxima* Michener 82. *meliloti* (Cockerell) 83. *aridula* Cockerell 84. *californica* (Ashmead) 85. *opuntiae* (Cockerell)

TABLE 1—*Continued*

Genus group	Genus	Subgenus	Species group	Species code number and name
		Titusella	—	86. *cubiceps* (Cockerell)
		Rhamphorhina	A	87. *foxiella* Michener
			B	88. *timberlakei* Michener 89. *eurynorhyncha* Michener 90. *stenognatha* Michener 91. *salviae* Michener 92. *cazieri* Michener 93. *clypeodentata* Michener 94. *australis* (Cockerell)
		Corythochila	—	95. *breviceps* Michener 96. *inyoensis* Michener
		Cubitognatha	—	97. *xenomastax* Michener

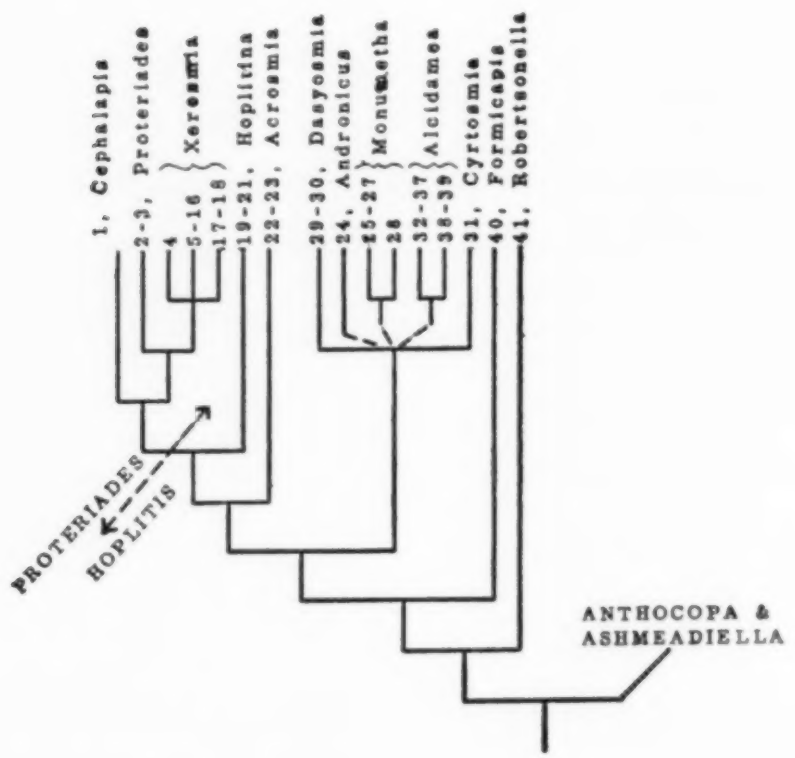

FIG. 1. Diagram of relationships for species groups in the genera *Proteriades* and *Hoplitis* as visualized by C. D. M. at beginning of this study.

The diagram has been modified from the more conventional dendritic form in order to be comparable to later diagrams (especially figs. 5–8 and 12–15). Relative levels of forks on any one main branch are related to primitiveness of the forms concerned. In contrast with the later figures, however, length of the various segments of the lines and relative levels of forks on different lines are of no significance. Another difference between this and later diagrams is that the tip of each line represents a species group, not a species. This is because C. D. M. did not attempt to indicate the structure within species groups. The broken line terminated by arrows separates the two genera. Generic names are capitalized, subgeneric names are in lower case type. The numbers at the tips of the lines are species code numbers.

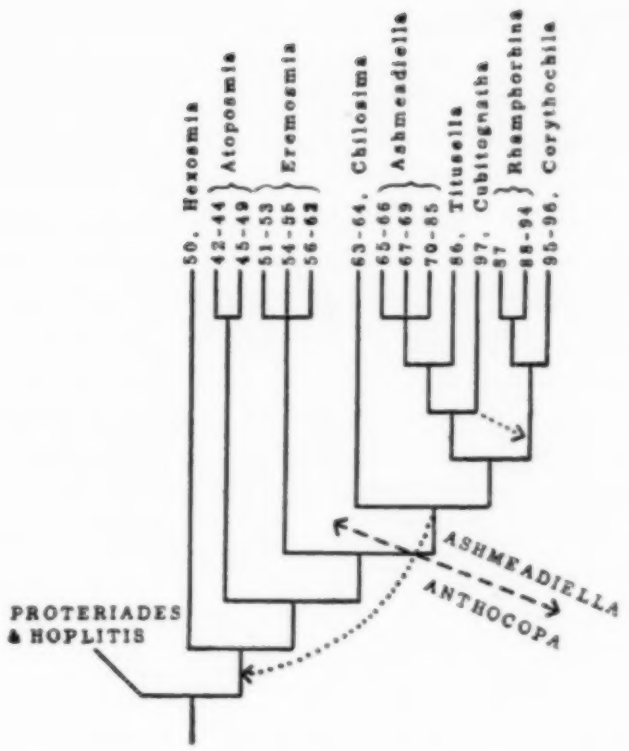

FIG. 2. Diagram of relationships for species groups in the genera *Anthocopa* and *Ashmeadiella* as visualized by C. D. M. at beginning of this study.

Explanatory comments as for figure 1. Dotted lines with single arrows point to possible alternative origins of certain stems.

significant changes in views being indicated in the latter paper.

These diagrams, constructed on the basis of systematic studies, undertaken and largely published before statistical work was contemplated, show the relationships as they were then understood. Since diagrams of relationships, phylogenetic trees and the like often have different meanings to different workers, we elaborate on the meaning of our diagrams as follows: They show that in the *Hoplitis* complex two major groups (1, *Proteriades, Hoplitis;* 2, *Anthocopa, Ashmeadiella*) arose from a common ancestor. The relative levels of forks on any one main branch are related to the primitiveness of the forms concerned. For example *Hexosmia* and *Robertsonella,* believed to have retained relatively primitive characters, should be more closely related than any other pair of subgenera in opposite major groups. Moreover, they should be more similar to the common ancestor than others in their major groups. The subgenera *Alcidamea, Andronicus, Cyrtosmia, Dasyosmia,* and *Monumetha* were believed equally related to one another. The location of any branch to the right or to the left is of no significance. For example, *Alcidamea* was thought to be as closely related to *Monumetha* as to *Cyrtosmia.* Lengths of the various segments of the lines, and relative levels of forks on different lines are of no significance.

In figures 1 and 2, as well as in table 1, and throughout the remainder of this paper, generic and subgeneric names are freely used to avoid needless lists of species names or numbers. However, in all aspects of the work every effort was made to avoid bias due to the previous existence of such names. In the interest of brevity, the species of bees will hereafter be referred to by number only.

Characters Studied

The characters utilized were not the result of a new search, but were those that had been found of value in distinguishing any American species or group within the *Hoplitis* complex. Not all of them had been used or described within any one genus, but all had been used somewhere within the complex.

The word "character" has been used in two rather different ways by taxonomists. In its commonest usage, any feature of one kind of organism that differentiates it from another kind is a character. Thus the red abdomen of one bee is a character distinguishing it from another bee with the abdomen black. In this paper we use the word in a second connotation only; that is, as a feature which varies from one kind of organism to another. Now, to use the above example, abdominal color is the character; it occurs in two "states" or alternatives, red and black. The way in which the states were delimited will be more clearly understood from a perusal of examples detailed in the following paragraphs.

A total of 122 characters were found, each varying among the species so that

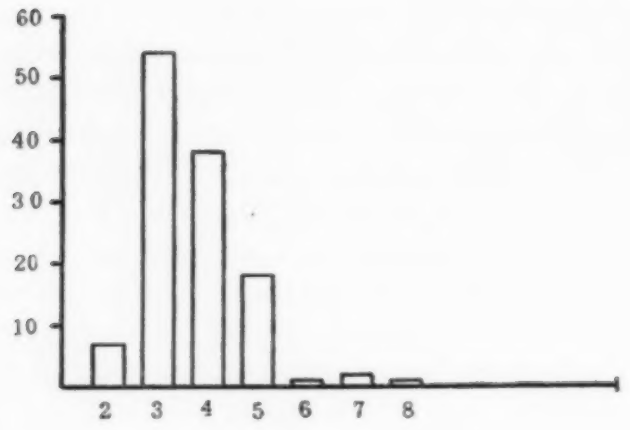

FIG. 3. Bar diagram showing number of states per character in the 122 characters of the study.

Abscissa: numbers of states per characters. *Ordinate:* frequency, number of characters per class. Average number of states per character equals 3.69.

there were from two to eight alternatives or states per character. Figure 3 is a bar diagram showing the number of states per character for the 122 characters of the study. The average number of states per character was 3.69. Numbers ranging from 1 to 8, hereafter called "state codes," were used to record the state of all characters for each species. The total number of character states was 450. Some characters could easily have been more finely subdivided to make more states.

In the study of the 97 species we considered a total of 11,834 characteristics. Each of the many characters that are regularly correlated between the sexes, even though different in males and females, was treated as a single character. Thus, the width of the genal area in relation to the eye was determined for females and ignored in males. It is usually markedly less in males than in females of any one species and is often used in distinguishing species in both sexes. It could reasonably have been used as two separate characters, one for each sex, each with several states. The number of characters could easily have been increased by about one-third had all similar cases been treated in this way.

Table 2 lists the characters studied, with the number of states into which each was subdivided. Reference to this table will show that characters of many sorts were used. The following paragraphs illustrate by means of examples the way in which we dealt with most of the problems encountered. Of course the states were always arranged in some logical linear sequence. A few characters were meristic, such as number of segments of the maxillary palpi, which ranges from 5 to 2, with obviously 4 states (state 1 = 5-segmented; 2, 4-segmented; 3, 3-segmented; 4, 2-segmented).

Sizes of the body and of various structures were used as characters. In most such cases a complete range existed from the largest to the smallest; the range was arbitrarily divided into a convenient number of states and appropriate states recorded for each species; when a species fell on a borderline between states, it was placed arbitrarily on one side or the other.

Coloration was treated in a similar manner. For example, the metasoma varies from red to black; 4 states were employed: (1) black, (2) red on the first and second terga only, (3) red on the first to third or fourth terga, and (4) metasomal terga entirely red or nearly so. A few species have a borderline type of coloration or their range of coloration overlaps two states and hence could have been placed, for example, in either 3 or 4. They had to be placed arbitrarily in one or the other. Fortunately we were not confronted with forms in which only the apical terga were red; had such existed, an effort would have been made to distinguish the two types of coloration for all species and to recognize two characters, one for red expanding from the base of the metasoma, the other for red expanding from the apex.

Red markings of the same sort occur in some species of each of the four genera. This is an obvious example of parallelism (the presumed primitive forms of each genus have little or no red) but we used the same character state numbers for the same condition in each genus. To do otherwise would introduce a deliberately

TABLE 2

Characters, with the number of states into which each was divided shown at right. Characters marked by an asterisk (*) were observed in females but are so well correlated with the same male structures that no separate characters are listed for the males. Characters marked by a dagger (†) are for males only. Those that are unmarked are for females only.

*Size	4
Extent of projection of apex of clypeus over labrum	4
Degree to which clypeal apex projects forward rather than downward	4
Breadth of clypeal truncation (broad to narrow and produced)	7
Ends of clypeal truncation (almost unrecognizable to much produced)	6
Shape of clypeal truncation (convex to concave)	4
Sublateral angles of clypeus	5
Emargination between truncation and sublateral angle of clypeus	5
Width of impunctate margin of clypeus	4
Convexity of clypeus	5
Spaces between clypeal punctures (close to clypeus impunctate)	4
Longitudinal median line of clypeus (absent to distinct and raised)	4
Coarseness of clypeal punctures (finer than to coarser than those of frons)	3
*Coarseness of punctures of vertex (finer than to coarser than those of mesoscutum)	3
*Ratio of distance from antennal bases to anterior ocellus to distance from anterior ocellus to posterior margin of vertex	4
*Ratio of distance between posterior ocelli to distance from one of them to eye margin	3
*Ratio of distance between posterior ocelli to distance from them to posterior margin of vertex	4
*Ratio of genal width to eye width	5
Degree of convergence (or divergence) of inner margins of eyes	3
Clypeal brushes (absent to large)	5
Mandibular hairs (sparse to dense brush)	4
Width of mandibular apex	4
Width of mandible at narrowest median region	4
Length of mandible	3
Size of upper mandibular tooth (absent to large)	3
Inner subapical swelling of mandible (absent to tooth-like)	4
Ratio of distance from upper to middle mandibular tooth to distance from middle to lower tooth	5
Shape of upper mandibular tooth (acute to absent)	4
*Height of hypostomal carinae	3
Posterior angles of hypostomal carinae	2
Shape of hypostomal areas (long to short)	3
Width of proboscidial fossa	3
*Segments of maxillary palpus (5 to 2)	4
*Ratio of length of first to second segment of labial palpus	4
*Setae on labial palpi and galeae (few straight, to many wavy)	5
Shape of labrum (broad rounded, to narrow bidentate)	4
*Length of proboscis	4
Ratio of length to breadth of antennal pedicel	2
Ratio of lengths of first to second flagellar segments	4
Ratio of length of antennal scape to distance between antennal bases	3
*Shape of thorax (elongate to nearly spherical)	4
*Pre-episternal carina (absent to long)	3
*Carina of pronotal lobe (absent to distinct)	3
*Hind tibial spur (slender to robust)	2
*Hind coxal carina (absent to high lamella)	5
*Coarseness of punctures of mesoscutum (by comparison to those of episterna)	3
*Coarseness of punctures of episterna (by comparison to those of genal areas)	3
*Spaces between mesoscutal punctures	4
*Pattern of pubescence at anterior end of mesoscutum (none, pair of spots, merged spots)	4
*Size of papillae on wings	3
*Wing color (clear to dusky)	3
*Depth and definition of concavity of first metasomal tergum	4
*Number of fasciae on metasomal terga	5
*Coarseness of largest metasomal punctures (by comparison to those of mesoscutum)	3
*Coarseness of metasomal punctures in general	4

TABLE 2—*Continued*

*Width of smooth margins of metasomal terga	3
*Distinctness of basal grooves of metasomal terga	2
Length of scopa	3
Amount of scopa on sixth metasomal sternum	3
Size of depression of sixth metasomal sternum (absent to broad)	4
Amount of projection of sixth metasomal sternum beyond corresponding tergum	3
Profile of sixth metasomal tergum (straight to strongly concave)	3
Width of apical flange of sixth metasomal tergum	3
*Integumental color (black to metallic)	3
*Metasomal color (black to red)	4
*Femoral color (black to red)	4
*Color of tegulae (black to red)	4
*Color of pubescence (white to black)	4
†Breadth of clypeal truncation	4
†Ends of clypeal truncation	4
†Shape of clypeal truncation (smooth to crenulate)	3
†Sublateral angles of clypeus	3
†Width of impunctate margin of clypeus	3
†Convexity of clypeus	4
†Spaces between clypeal punctures	3
†Coarseness of clypeal punctures (by comparison to those of frons)	3
†Direction of subantennal sutures (parallel to diverging upward)	3
†Degree of convergence (or divergence) of inner margins of eyes	3
†Density of clypeal vestiture	4
†Clypeal brushes (absent to distinct)	3
†Length of mandible	4
†Size of upper mandibular tooth (absent to large)	3
†Length of lower mandibular tooth in relation to others	3
†Apex of labrum (rounded to emarginate)	2
†Antennal scape (slender to swollen)	3
†Antennal apex (rounded, pointed, hooked, flattened)	4
†Length of hair of antennal flagellum	3
†Ratio of length to breadth of antennal pedicel	4
†Thickness of antennal flagellum	3
†Ratio of length to breadth of median flagellar segments	3
†Outer carina of hind coxa	2
†Ventral tooth of middle coxa	3
†Lateral teeth of sixth metasomal tergum (absent to acute)	4
†Median teeth of sixth tergum (absent to twice as long as broad)	5
†Apical flange of sixth tergum (absent to broad)	3
†Lateral margins of sixth tergum (straight to convex)	3
†Extent that seventh tergum is exserted	5
†Shape of apex of seventh tergum (medially pointed, rounded, to bilobed)	7
†Profile of first metasomal sternum (flat to protuberant)	3
†Size of second sternum in relation to third	3
†Profile of disc of second sternum (flat to protuberant)	3
†Apex of second sternum (flat to transversely elevated)	3
†Margin of second sternum (concave to medially pointed)	5
†Margin of third sternum (concave to convex)	5
†Fringe of third sternum (absent to long across entire margin)	5
†Margin of fourth sternum (concave to convex)	4
†Fringe of fourth sternum (absent to long)	3
†Amount that fifth sternum is exposed	3
†Margin of fifth sternum (concave to convex)	3
†Fringe of fifth sternum (absent to long)	3
†Amount that sixth sternum is exposed	3
†Longitudinal ridge of sixth sternum (absent to produced and expanded)	5
†Sixth sternum (planate to duplicated)	3
†Margin of sixth sternum (convex, to concave, to concave with median point)	5
†Subapical lobes of sixth sternum (present or absent)	2
†Margin of seventh sternum (convex to concave)	5

TABLE 2—*Continued*

†Projection of eighth sternum behind lateral apodemes (absent to over twice as long as broad)	5
†Curvature of gonocoxites (none to angular)	3
†Apices of gonocoxites (pointed to truncate)	3
†Shape of volsellae (short rounded, to long pointed)	3
†Fusion of volsellae to gonocoxites (none to complete)	3
*Distribution (desert only to high montane only)	8

subjective element and moreover, as explained elsewhere, the large number of characters used swamps the effects of occasional parallelisms.

Another type of coloration used is intensity of blue or green metallic hue. This is entirely independent of the red coloration; actually no species exist in the *Hoplitis* complex which are both red and metallic but this combination occurs in related bees and is presumably not physiologically impossible in the *Hoplitis* complex. On the other hand, we excluded color of the red pigment as a character because, had we used it, one state (lack of red) would have been the same for two characters (extent of red and color of red pigment). In all cases characters were reorganized or omitted to exclude correlations resulting from such situations, where a certain state of one character requires a particular state of another.

Proportions of various structures were widely used. For example, the ratio of the first to the second segments of the labial palpi was divided into four states as follows: greater than 1.0, 1.0 to .75, .74 to .5, less than .5. As with other continuously variable characters, various species fell on the border lines between states or the range of their variation overlapped two states; such species were placed arbitrarily in one or the other of the two states. No effort was made to establish character states that would contain equal numbers of species; actually, in this instance, there are few species in the first and last states and many in the two middle states. Rather, in so far as possible, character states with median points more or less equally different morphologically were employed. Characters of sculpturing (punctation) of the body were handled in a similar manner. For example, clypeal punctation of the males was divided into four states, much finer than that of the frons, finer than the frons, equal to the frons, and coarser than the frons. In such cases, to insure uniformity of judgment, a certain species was selected in advance as typical of "much finer than frons," another as typical of "finer than frons."

Among some of the structural characteristics, such as form of the clypeus of the female, were encountered the only problems in arranging character states in a logical order. In this instance the difficulties disappeared when the clypeal form was broken down into several component elements as shown in the first part of table 2.

No character was included, of course, if it was the same in all species of the *Hoplitis* complex. However, characters were included in cases where only a single species differed from all the rest. There are several such instances of which the following exemplify two types: The inner subapical swelling of the mandible of the female is absent (state 1) in the majority of species, present and rounded or weakly angulate (state 2) in many *Anthocopa*, and produced to a distinct tooth (state 3) in but one species. An outer carina on the hind coxa is absent (state 1) except in one species (which exhibits, therefore, state 2).

After the data were collected, it was apparent that C.D.M. had had a strong tendency to begin the series of states for the various characters with number 1 as the commonest extreme or the extreme state nearest the commonest state. The consequences of this fact and of the variable number of states per character are

discussed in the section on "Choice of a Correlation Coefficient" by Sokal and Michener (1957).

Procedures

Correlational Procedures

As already indicated, the justification for and details of our statistical procedures are being presented elsewhere (Sokal and Michener, 1957). Here we offer merely a summary statement.

Two obvious ways suggested themselves for condensation and interpretation of the data discussed above. We could either correlate characters with each other (R-technique of factor analysts) or species with each other (Q-technique of factor analysts). The former is the usual technique in biological and psychological studies involving correlational analysis. For our purposes, however, correlations between species based on the characters studied, seems a more useful method. In so far as the characters used are indicative of the entire spectrum of potential variation of the species, we can say that the resulting correlation coefficient is representative of the real affinity between two species. When scanned for clusters of high correlation coefficients, the Q-type matrix (i.e., tabulation of species against species showing correlation coefficients of each species with all of the others) reveals species which are similar. It is thus especially suited to classificatory problems. In spite of this, except for the work of Sturtevant (1942) which involved not correlations but character differences, the authors are unaware of other Q-type studies in the biological sciences.

There are two evolutionary situations under which it is important to examine the two types of matrices. The first might be referred to as breakage of correlation. It occurs when two characters that were correlated in the ancestral line and are still correlated in related lines become independent of each other in a certain evolutionary branch. Under such conditions the R-matrix is a poor representation of the true relation between the two characters. There is no good way of representing such a correlation, close in one line, absent in the other. On the other hand a Q-matrix is not affected by such data.

Convergence of species for a number of characters is a second disturbing phenomenon. Here the R-matrix is not affected while the Q-matrix is affected if the convergent characters outweigh the non-convergent ones in numbers. As explained in the Introduction, we do not believe this is likely if an adequate number of characters is studied. In case of a preponderance of convergent characters and in the absence of paleontological data it is doubtful whether the systematist would be able to distinguish convergence from relationship by descent.

After considering the various possibilities, the product-moment correlation coefficient was adopted as the most suitable, as explained by Sokal and Michener (1957). It might have been desirable to code the data in such a way as to put all character states on the same scale. In a character with two states, the code "2" indicates one extreme for the character, quite unlike code "2" in a character, with, for example, 7 states. This situation is usually met by normalizing rows (characters) of the raw data. We did not perform this transformation for various reasons, including the prohibitive amount of IBM (International Business Machines) work that would have been necessary, since a one-digit code would not have sufficed for normalized data.

The authors are well aware that their methodology of coding and correlation could profit by refinement. It is, however, our point of view that in a pilot study of this nature such refinements are premature. Should the general method prove of value, significant results will surely emerge in spite of minor imperfections in technique.

Even though simplified by the use of a one digit code rather than actual meas-

TABLE 3. *Two-way frequency distribution of correlation coefficients against formal classification codes*

r-class code	r	Formal classification code (c) 1	2	3	4	5	Total f_r
1	−07					5	5
2	−02					20	20
3	03				3	36	39
4	08				9	46	55
5	13				20	79	99
6	18			1	27	162	190
7	23			5	51	298	354
8	28			5	140	384	529
9	33			7	150	431	588
10	38			33	181	440	654
11	43			86	189	275	550
12	48		2	121	141	92	356
13	53		13	154	88	22	277
14	58	1	10	120	73	6	210
15	63	5	17	94	59		175
16	68	14	27	53	10		104
17	73	22	32	32	4		90
18	78	63	49	17	4		133
19	83	87	36	4			127
20	88	57	16				73
21	93	23					23
22	98	5					5
Total f_c		277	202	732	1149	2296	4656

urements or any other device to express character states, the computation of a 97 species × 97 species matrix of correlation coefficients presents serious technical difficulties. An operation of this magnitude cannot be reasonably undertaken without punched card or electronic computing equipment.

The Matrix of Correlation Coefficients

The 4656 correlation coefficients among the species of our study ranged in magnitude from − .0626 for the correlation between species 26 and 92, to .9747 for the correlation between species 43 and 44.[3] Correlation coefficients equal to or larger than .23 are significant at the one per cent level. As our next procedure, a frequency distribution of these correlation coefficients was set up with the help of the IBM tabulator. The correlation coefficients were grouped into 22 classes with class intervals of .05. The frequency distribution can be read from the right hand column of table 3. The modal class has a class mark of .38; this represents the most frequent class of correlation coefficients found between species in this study. However there is a second mode at r = .78. This bimodality would indicate that we are dealing with two populations of correlation coefficients: Those indicating close, possibly intrageneric relations and others representing more distant relations. That this is so can be verified from a two-way frequency distribution of the formal classification against the correlation coefficients, shown in the body of table 3. Codes across the top of the table represent relations between species based on the formal classification as

[3] For lack of space the matrix cannot be reproduced here. Microfilm or IBM-tape or card copies can be obtained through the Secretary, Department of Entomology, University of Kansas, Lawrence.

follows:

1. Species in the same species group.
2. Species in different species groups but in the same subgenus.
3. Species in different subgenera but in the same genus.
4. Species in different genera but in the same genus group.
5. Species in different genus groups.

It is clear that a large part of the second peak is formed by relations coded 1, i.e., between members of the same species group. It was encouraging to find that magnitude of the correlation coefficient was apparently an estimate of systematic relationship as indicated by the formal classification. This was confirmed by a calculation of the correlation coefficient between the computed correlation coefficients and the codes for the formal classification which was − .80.

Another way of examining these correlation coefficients is to study frequency distributions of the coefficients for any single species against all other species. Figure 4 shows histograms of such distributions for species 1, 4, 6, 19, 29 and 92. There are marked differences in form between the histograms. Species 1, 4, 6, 19 and 29 have two modes and species 92 has three modes. The principal modes are at .33 for species 1, 19 and 92 and at .38 for species 4, 6 and 29. The secondary mode is highest in species 6 (.78), lower in 1, 4 and 19, and lowest in 92 (.53). This latter species has a third mode at .86. The scatter is lowest in species 1 and 19, increased in species 4, 6 and 29 and highest in 92.

On the basis of these findings we can distinguish at least three types of species. The first type is a member of a closely related group of species. It therefore has a number of correlations considerably higher than the majority of its correlations. Species 6 and 92 with their high secondary modes are examples of this type and subsequent work has borne out this view. Close inspection reveals differences between the two. Species 92 has three distinct modes, while species 6 has only two with a slight indication of a third one. Subsequent studies showed that species 92 was a member of a closely knit species group with a considerable gap between this group and the rest of its genus (see fig. 8). Species 6 is also a member of a species group but transition to more distant relations in the same genus is more gradual than in the former case (see fig. 5). It is interesting to note the tail of low correlations shown by species 92, while the correlations of species

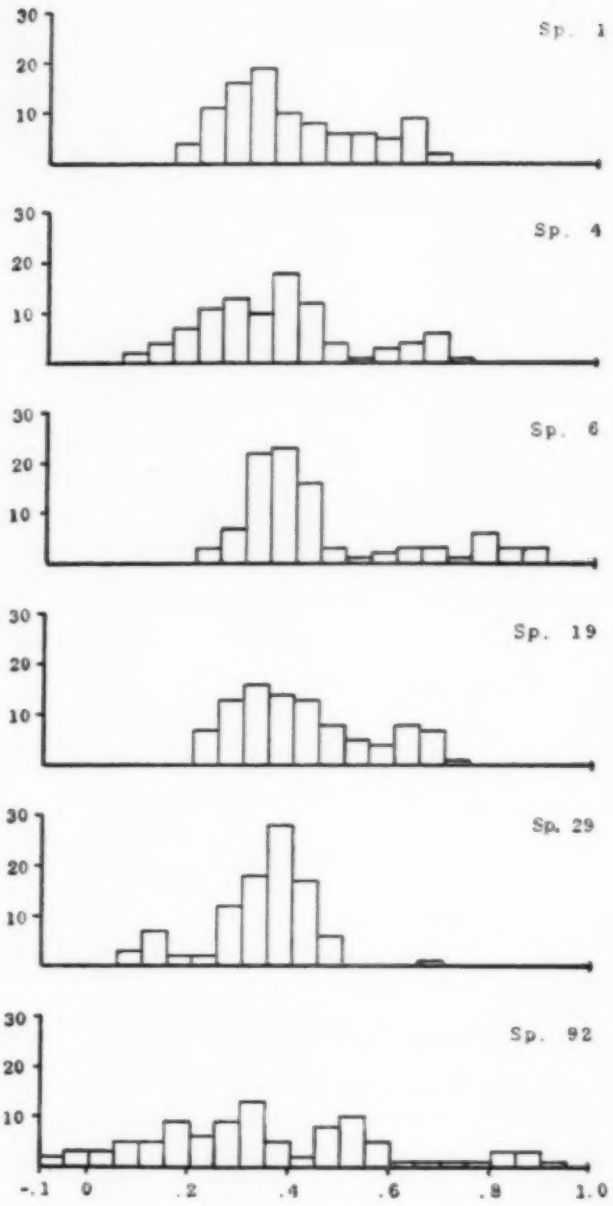

FIG. 4. Histograms showing levels of correlation coefficients for each of six species against all other species.

Abscissas: levels of r for given species. *Ordinates:* frequency, number of r's per class.

6 are sharply truncated at the lower end. It would appear that species 6 is closer to the general type of the *Hoplitis* complex than is species 92.

The second type of species is illustrated by 1, 4 and 19. They are isolated species moderately well correlated with the other members of their genus, as illustrated by their lower secondary modes. Species 4, by its greater scatter, appears to deviate more from the norm than the other two. Reference to figure 5 will confirm these views.

The third type is the very isolated species, which is possibly in a genus by itself. The histogram of species 29 shows only one relatively high correlation (with species 30 at .60). The remaining correlations are far below that value. Our subsequent study suggested that 29 and 30 may be different enough from the other species in the *Hoplitis* complex to warrant generic distinction (see fig. 6).

The absence of significant negative correlations from our matrix requires some discussion. Q-technique matrices of correlations between people (based on psychological tests) are quite likely to yield such correlations. If there are distinct, antithetical types of persons represented in the matrix, such as extroverts and introverts, it is likely that a high score for one type will be a low score for the other and vice versa. In our case evolutionary progress may be represented by either an increase or a decrease in state codes. In the majority of characters the supposedly primitive situation is an intermediate state code with two diverging evolutionary trends represented by the lower and higher code numbers. Furthermore, characters representing correlated trends were not necessarily coded on the same scale. It is clear that under such circumstances distantly related forms are likely to be uncorrelated rather than negatively correlated.

The Search for Group Structure

The matrix of correlation coefficients obtained as reported above can be put to a variety of uses and the analysis reported below represents merely an initial effort at exploitation of the data. The correlation coefficients serve as an absolute measure of relationship between any two species in our study, limited only insofar as the characters chosen do not represent the total correlated variation of the two species. Serious study of the matrix will center around two aspects: (1) a search for group structure (2) an analysis of the evolutionary factors in the systematic group under study. The second point will undoubtedly require some method involving the use of partial regressions, be this factor analysis, path coefficients or some other device. This study is presently being initiated and the authors are optimistic about the significance of the outcome (see Sokal, 1957).

The search for structure among the correlation coefficients of the matrix is of course no different in aim from the search by the systematist for a natural system in an array of species. Such a system consists of a hierarchy of groups. Various methods can be used for discovering a hierarchy in data such as ours. A customary, rather simple device of the psychometrician is so-called "cluster analysis," developed to a fine art by Tryon (1939).

Attempts to employ cluster analysis for finding structure in our matrix were only partially successful, since the resulting clusters were partly overlapping, i.e., a given species might be a simultaneous member of two clusters. This makes good sense for intermediate forms in an abstract scheme of relationships. In a systematic hierarchic classification, however, groups at the same level have to be mutually exclusive for practical as well as for theoretical reasons, except for low level groups exhibiting reticulate evolutionary patterns (rare above the species level in animals). A further reason for the unsuitability of cluster analysis is the complexity of the clusters as more species are added to them. Although clusters are therefore not convenient in an initial

search for structure, the diagram of relationships established by methods to be described below could be easily recognized in the clusters outlined by cluster analysis. A method essentially similar to cluster analysis is the ρ-group and ρF-group method of Olson and Miller (1951) applied to three paleontological R-technique matrices. It suffers from the same drawbacks as cluster analysis.

In view of the disadvantages of this and certain other methods, we decided on the following procedure: a nucleus of a group was established, using the two species having the highest coefficient of correlation. Then species would be added to this nucleus, one at a time, always adding first the species with the highest average correlation with the members of the group. The limit of the groups could be found by decreases $(\bar{L}_{n+1} - \bar{L}_n)$ in the level of the average correlation L_n, where the subscript refers to the number of members in the group. A significant drop was empirically determined since sampling distributions of average correlations,

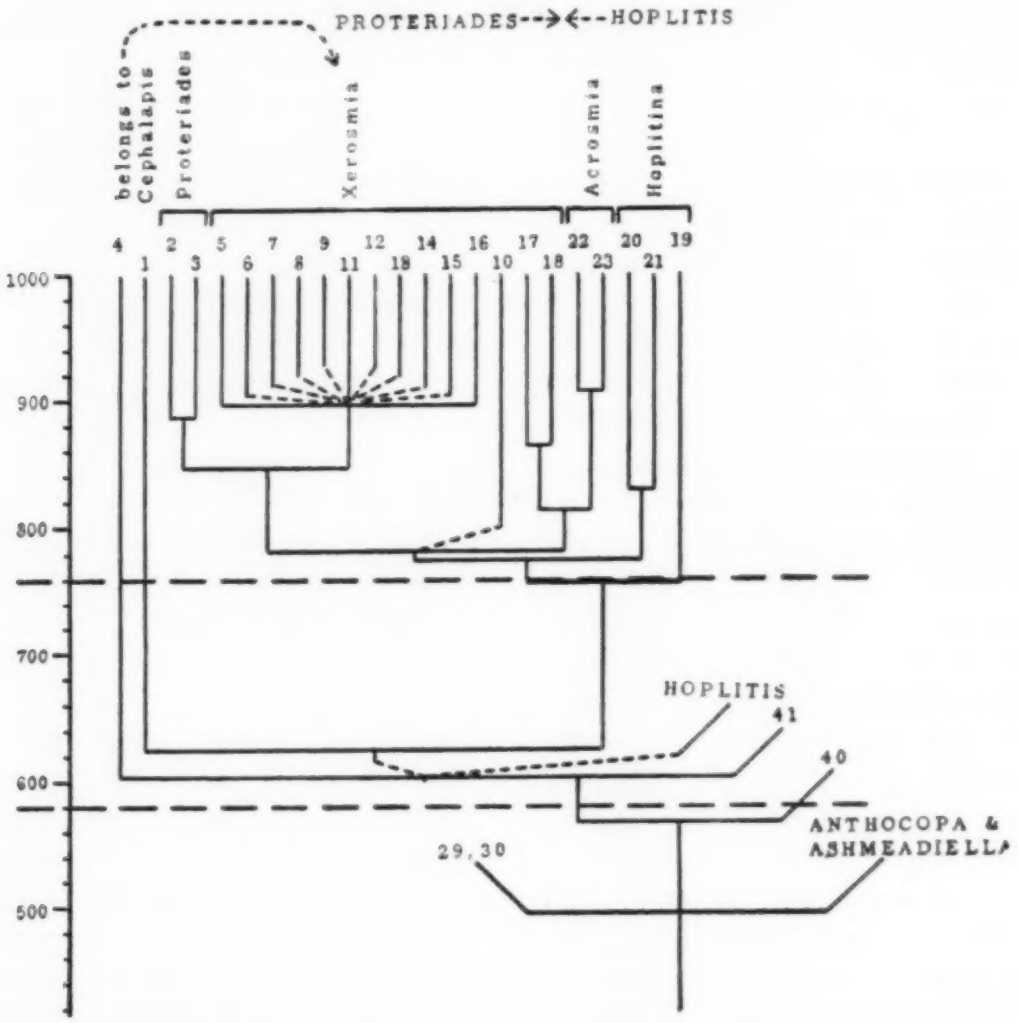

FIG. 5. Diagram of relationships for the genus *Proteriades* obtained by the weighted variable group method.

Ordinate: magnitude of correlation coefficient multiplied by one thousand. Correlations between any two joining stems can be found by reading the value on the ordinate corresponding to the horizontal line connecting the stems. This value becomes approximate and maximal in cases of multifid furcations. Broken lines where more than 2 lines join, are for convenience only; the horizontal connecting line has the same significance as elsewhere. "Roofs" over the species numbers at the tips of the lines delineate subgenera containing more than one species. These subgenera, identified in lower case print, are based on C. D. M.'s formal classification and not on the findings of this study. Generic names are capitalized. The boundary between the genera *Proteriades* and *Hoplitis* according to the older classification is indicated by two arrows meeting head-on at the top of the diagram. Horizontal broken lines are explained in the text, under Classificatory Problems.

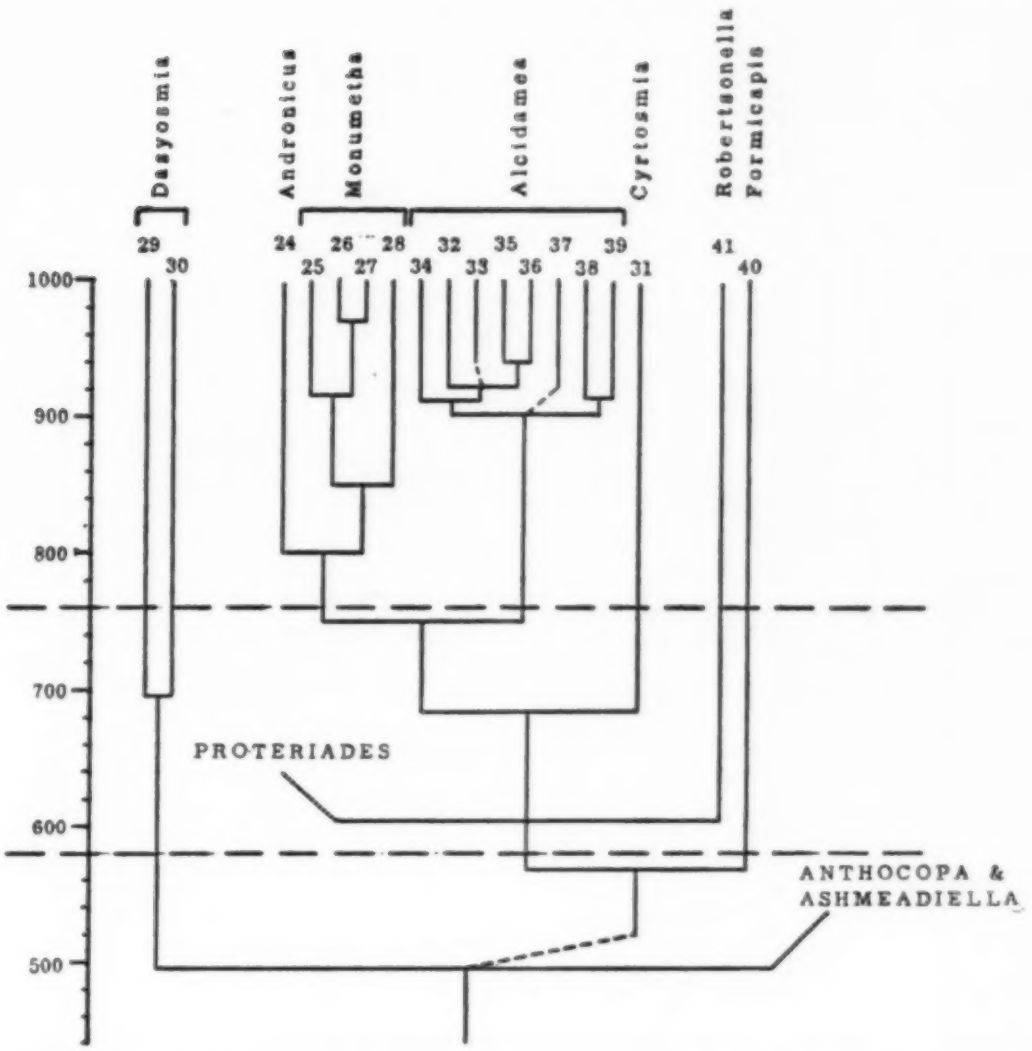

FIG. 6. Diagram of relationships for the genus *Hoplitis* obtained by the weighted variable group method.

Explanatory comments as for Figure 5.

such as $\bar{L}_n$, are unknown. First, lower groups (species groups) were developed. Then a new matrix of correlation coefficients, based on the groups as well as isolated species which had failed to enter any group, was calculated. Then by the same method, it was possible to unite these into larger groups (sometimes subgenera), and these into still larger or higher groups, etc. Thus it has been possible to develop a hierarchy of groups which is represented below by means of diagrams of relationships (figs. 5–8) for comparison with figures 1 and 2.

We might have admitted only one new member for each group at a given hierarchic level, thus obtaining a diagram of relationships consisting of bifurcations only (pair group method). However, we decided that this was an artificial situation and, moreover, we are forced into multifid furcations by some decreases in the average correlation coefficients, too small to plot. Thus a method using groups of various sizes, called a variable group method, was selected as more reasonable and practical than the pair group method. A second consideration was how to weight the variables (species or groups) on calculating a new correlation matrix following each grouping procedure. If *A* and *B* represent closely related species, *C* a more distant species, the problem is, should a still more distant species, *D*, be placed in relation to a homogeneous group *ABC* or in relation to the *AB-C* stem of a diagram of relationships? The latter alternative was selected, so that not all species have the same weight in constructing the diagrams

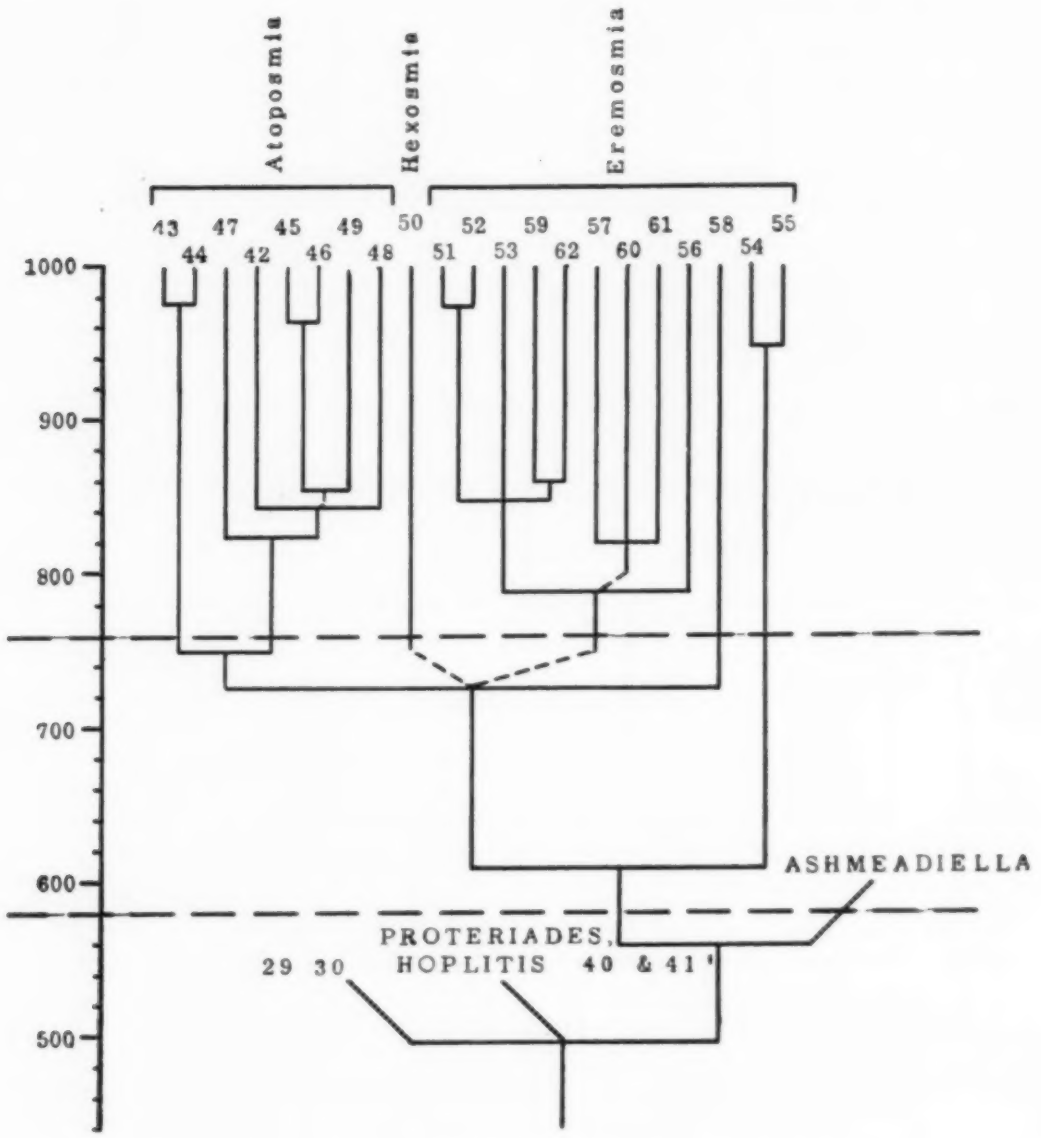

FIG. 7. Diagram of relationships for the genus *Anthocopa* obtained by the weighted variable group method.

Explanatory comments as for Figure 5.

of relationships. We therefore termed our technique the "weighted variable group method." The greater part of another paper (Sokal and Michener, 1957) is devoted to a detailed explanation of this method so that others can readily follow it.

Once computed, the relations were represented as diagrams of relationships in figures 5–8, one diagram for each genus. The ordinate at the left of each diagram is graduated in units of 1000 times the correlation coefficient. The correlations between any joining stems in the diagram can be read by measuring the level along the ordinate of the horizontal line connecting the stems. Thus in figure 8, species 63 and 64 are correlated at a level of .908, while group 63–64 is related to group 67–72 at .702. Furcations involving more than three lines are shown by broken lines converging on the midpoint of the horizontal line as in group 88–94 of the above figure. This was done to avoid any impression that, for example, 88 and 89 are closer than 88 and 94. The tops of the figures are at a level of 1000 (correlation of 1) since obviously each species is perfectly correlated with itself. These diagrams represent relationships in a static, non-historical sense.

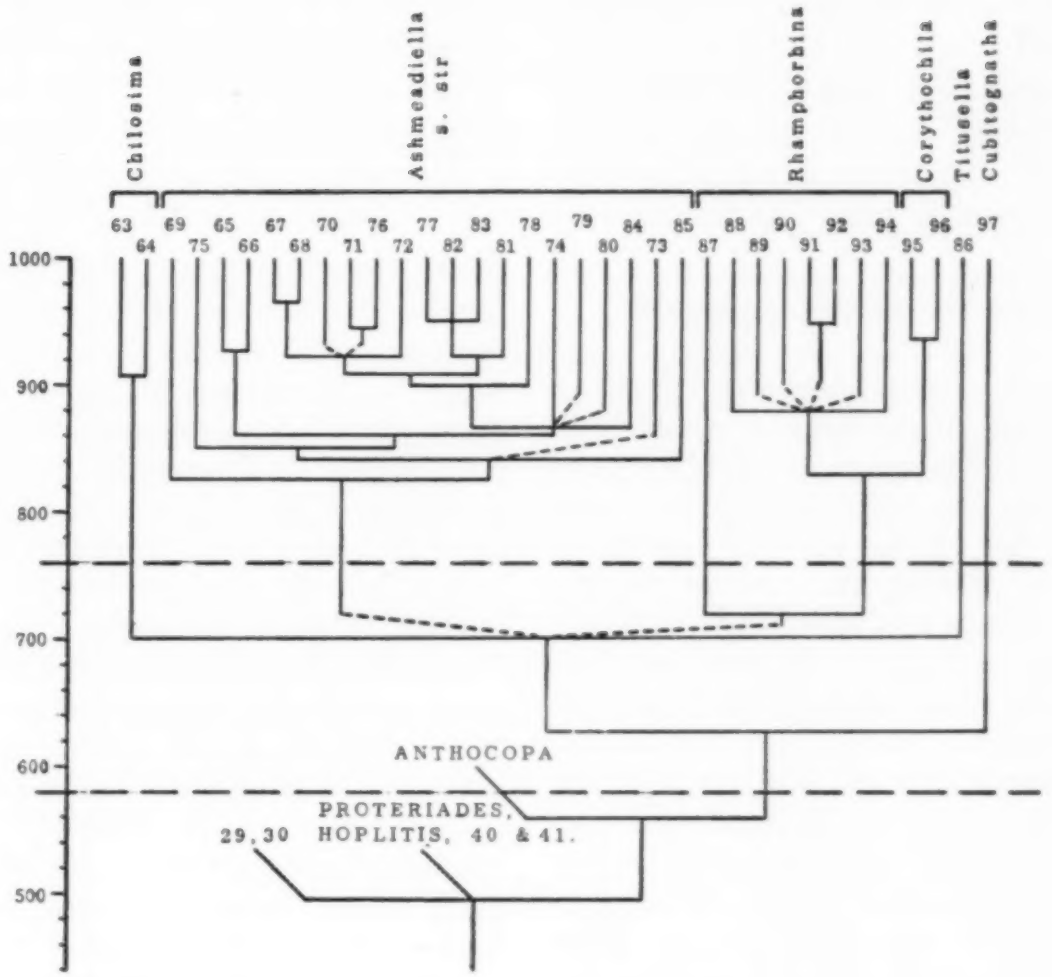

FIG. 8. Diagram of relationships for the genus *Ashmeadiella* obtained by the variable group method.

Explanatory comments as for figure 5.

COMPARISON OF DIAGRAMS OF RELATIONSHIP

Relationships

A comparison of table 1 or figures 1 and 2, the older classifications (by C.D.M.), with figures 5–8 shows that the correspondence between the classical and the quantitative methods is good, with certain exceptions discussed below. The diagrams of relationships shown in figures 5–8 differ from those of figures 1 and 2 in that the vertical distances indicate degree of relation (or of divergence) as shown by the scale of average correlations ($\times$ 1000) at the left. Thus the degree of difference between any two forms can be judged by measuring from the top of the figure to the level where their branches meet.

The following is a list of the main changes indicated by the new diagrams (figs. 5–8) upon comparison with the old ones (figs. 1–2):

1) 17–18 closer to 22–23 than to 5–16.
2) 19 a separate group from 20–21.
3) 1 and 4 more isolated from *Proteriades* in general.
4) New boundary between *Proteriades* and *Hoplitis*, the subgenera *Hoplitina* and *Acrosmia* being transferred to *Proteriades*.
5) More structure in junction of *Dasyosmia-Cyrtosmia* group of *Hoplitis*.
6) *Dasyosmia* extremely isolated.
7) 40 and 41 in reversed positions in relation to the main stem of *Hoplitis*.

8) 54 and 55, 86, and 97 more isolated.
9) More (and sometimes different) structure in *Monumetha, Alcidamea, Atoposmia, Eremosmia, Ashmeadiella* s. str., and *Rhamphorhina.*
10) 50 not as isolated as claimed.

There are several instances (listed in item 9 above) of groups not or but little subdivided in figures 1 and 2 which now show more structure; that is, we now have evidence of the varying degrees of relationship among the species of the groups. In part this is merely due to the fact that in the original diagram no effort was made to show relationships below the level of species group. In *Monumetha* the relationships shown in figure 9 are the same as those that would have been suggested had a "tree" for the species of this group been attempted before the statistical study. In the other groups, usual systematic methods did not provide the worker (C.D.M.) with the bases for attempting diagrams of relationships. Certain species (e.g., 67, 68) were known to be closely related, so that it is not true that these groups were "structureless" in the eyes of the systematist, but it was not obvious how the pairs and little groups of very similar species, together with the more isolated species, were related. It is also true that the three groups recognized in *Ashmeadiella* s. str. (fig. 2) were very informal and the failure of the statistical method to support them is not surprising to C.D.M. In short, the diagrams for the groups listed in item 9 above, except for certain matters in *Eremosmia* discussed later, seem satisfactory and shed new light on some of the relationships. The same may be said also of the relationships between the subgenera *Dasyosmia, Andronicus, Monumetha, Alcidamea,* and *Cyrtosmia* (item 5 of above list), shown as equally related in figure 2. Figure 6 provides an idea of the relationships of these groups which seems acceptable to C.D.M. The most striking fact here is the isolated position of *Dasyosmia.*

The new location of the dividing line between *Proteriades* and *Hoplitis* (item 4, above list) is not at all surprising. The old diagram of relationships (fig. 1) shows *Proteriades* as a mere offshoot of *Hoplitis,* having close relationships (pointed out by Hurd and Michener, 1955) to *Hoplitina* and *Acrosmia,* which were placed as subgenera of *Hoplitis. Proteriades* was separated as a rather large group having specialized hairs on the galeae and labial palpi for extracting pollen from the slender flowers of *Cryptantha.* This was a striking and convenient character and was correlated with some others, but the discovery of males of 17 and 18, which nearly or entirely lack such hairs, already weakened it.

There are several instances where a species or small group is shown as very much more isolated by our statistical method than by the classical methods of systematics. This is noted in items 3, 6, and 8 of the above list and item 2 may represent the same phenomenon. We have selected species 4 as a basis for further discussion. There is no doubt that this is indeed a strange insect and that it differs by many characters from its relatives. However, one of us (C.D.M.) maintains that in spite of its numerous unique characters it shows definite evidence of relationship to species 1–16 and that it did not branch from the stock ancestral to all the other *Proteriades* as our method (fig. 5) might suggest. The bases for this contention are that species 4, like the rest of the group 1–16, has the following characters: (1) proboscis unusually short; (2) galeae and labial palpi provided with hooked hairs in both sexes; (3) maxillary palpi short; (4) middle trochanter of the male provided with toothlike lamella; and (5) the hind coxae provided with a strong carina. That this combination of characters arose independently, in parallel fashion, for species 4 and the other species from 1–16 is possible,

but unlikely. Of course the characters listed above are only some of those in which species 4 agrees with the others mentioned; numbers (1), (2) and (4) are peculiarly significant because they do not occur in the *Hoplitis* complex except among the species 1–16.

Our method shows species 4 to be quite distantly related to other members of the *Hoplitis* complex. However, the method cannot indicate whether a species differing greatly from others does so because it is a *pregroup* derivative, i.e., a form which has branched from the ancestral stem of a group; or because it is an *exgroup* species, i.e., a highly specialized derivative of the group in question with many features peculiar to it. The difference between these conditions is shown diagrammatically by phylogenetic trees in figure 9. In figure 9A, species X is shown as a derivative of a primitive stem; in 9B species X is a specialized member of a cluster of related species. The correlation coefficients between X and members of the cluster in 9A and between X and members of the cluster in 9B might be equally low. Yet the phylogenetic relationships are very

TABLE 4. *Relations among relatives of isolated species*

Isolated species		Relative number: 1	2	3	4	5	6	7	8	9	10	11	12
1.	Relative species	2	14	8	15	13	9	17	11	16	7	5	3
	r_{1j}	.69	.69	.65	.65	.64	.63	.62	.62	.61	.61	.61	.60
	baseline		.85	→	→	→	→	.78	→	→	→	→	→
4.	Rel. sp.	2	6	8	14	15	16	9	5	11	13	12	7
	r_{4j}	.71	.70	.70	.69	.69	.67	.67	.65	.65	.63	.62	.60
	baseline		.85	→	→	→	→	→	→	→	→	→	→
19.	Rel. sp.	17	5	22	14	20	23	18	11	6	13	16	12
	r_{19j}	.74	.69	.68	.68	.68	.68	.67	.66	.65	.65	.65	.64
	baseline		.78	→	→	→	→	→	→	→	→	→	→
29.	Rel. sp.	30	24	27	34	35	36	52	38	59	62	25	32
	r_{29j}	.70	.49	.49	.47	.46	.46	.46	.45	.45	.45	.44	.44
	baseline		.50	→	→	→	→	→	→	→	→	→	→
31.	Rel. sp.	27	25	26	37	28	32	24	34	35	33	36	38
	r_{31j}	.70	.69	.68	.62	.61	.60	.58	.57	.56	.56	.56	.56
	baseline		.92	→	.75	→	→	→	→	→	→	→	→
35.	Rel. sp.	36	32	33	34	37	39	38	27	25	24	26	28
	r_{35j}	.94	.93	.92	.89	.85	.84	.83	.63	.61	.60	.60	.58
	baseline		.92	→	.91	.90	→	→	.75	→	→	→	→
40.	Rel. sp.	28	25	26	27	17	37	50	41	94	18	1	24
	r_{40j}	.61	.58	.57	.53	.52	.52	.51	.50	.49	.49	.48	.47
	baseline		.85	→	→	.61	→	.50	→	→	→	→	→
41.	Rel. sp.	25	19	18	17	27	39	34	28	35	5	26	33
	r_{41j}	.60	.58	.55	.55	.55	.54	.53	.53	.52	.52	.52	.52
	baseline		.61	→	→	→	→	→	→	→	→	→	→
43.	Rel. sp.	44	42	45	46	49	48	47	61	51	60	53	52
	r_{43j}	.97	.80	.73	.72	.67	.66	.64	.61	.59	.58	.57	.55
	baseline			.84	→	→	→	→	.72	→	→	→	→

TABLE 4—*Continued*

Isolated species		Relative number: 1	2	3	4	5	6	7	8	9	10	11	12
50.	Rel. sp.	51	48	58	84	52	45	59	46	80	47	74	77
	r_{50j}	.73	.72	.70	.68	.66	.65	.65	.64	.64	.63	.62	.61
	baseline		.72	→	.56	→							
54.	Rel. sp.	55	80	77	85	82	73	79	59	72	58	75	81
	r_{54j}	.95	.65	.64	.63	.62	.62	.62	.61	.61	.60	.59	.59
	baseline			.56	→								
58.	Rel. sp.	52	51	50	59	62	53	56	61	57	60	54	80
	r_{58j}	.76	.74	.70	.69	.68	.67	.63	.61	.61	.60	.60	.59
	baseline		.98	.73	→							.61	.56
63.	Rel. sp.	64	70	96	76	67	71	68	95	78	72	65	80
	r_{63j}	.91	.75	.70	.70	.69	.68	.68	.68	.68	.67	.65	.64
	baseline			.70	→								
69.	Rel. sp.	72	68	85	82	81	67	70	75	79	77	83	76
	r_{69j}	.82	.82	.80	.79	.78	.77	.77	.77	.77	.76	.74	.74
	baseline		.92	.84	→								
86.	Rel. sp.	84	74	83	80	77	79	71	81	67	78	73	76
	r_{86j}	.75	.75	.70	.69	.67	.66	.65	.64	.64	.64	.64	.63
	baseline		.86	→								.84	→
97.	Rel. sp.	92	89	91	63	64	90	93	96	72	88	95	65
	r_{97j}	.66	.63	.61	.60	.60	.60	.59	.59	.58	.58	.58	.58
	baseline		.88	→	.70	→							

different. For convenience we refer to this problem as the "pregroup-exgroup problem."

A consideration of this matter and of the correlation coefficients among species of the *Hoplitis* complex suggests that quantitative ways of solving this dilemma may exist for particular species. An array of species most closely related to a given isolated species can be listed in order of decreasing magnitude of their correlation coefficients with that species. The "base line" relating these relatives to each other can be read off figures 5 to 8 as the highest horizontal line connecting the species concerned. Clearly, as more species are admitted to the group of relatives the base line level is bound to decrease. The original level of the base line, the presence or absence of a decrease and its magnitude appear to reveal phylogenetic relationships of the isolated species. For purposes of analysis we have chosen an arbitrary number of 12 nearest relatives for each of 16 isolated species and listed them in decreasing order of correlation with the isolated species in table 4. Line 1 for each isolated species

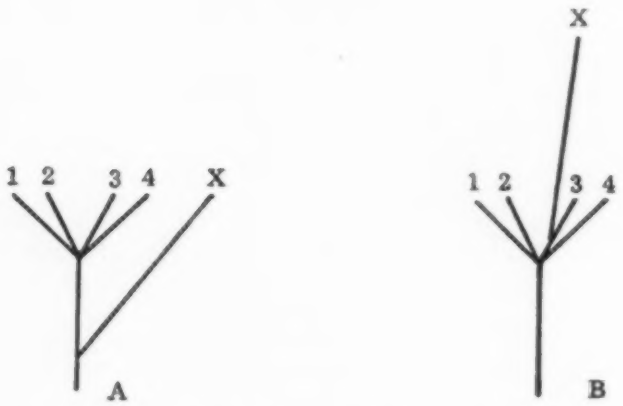

FIG. 9. Hypothetical diagrams of relationships to illustrate the problem of differentiating between primitive and specialized differences, or pregroup and exgroup species.

For explanation see test.

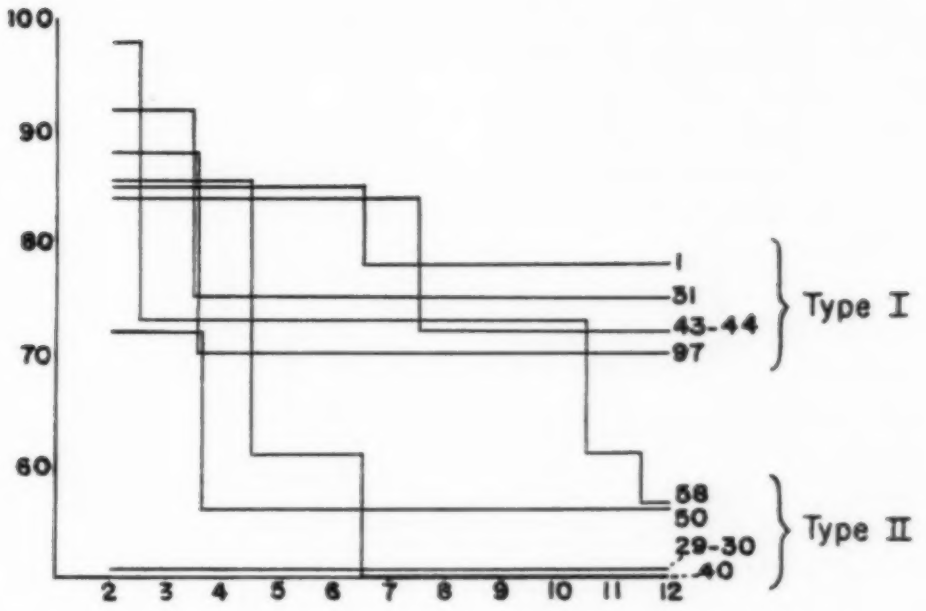

FIG. 10. Base lines of relatives of isolated species types I and II (data from table 4).
Abscissa represents decreasing rank of correlation of isolated species with related species. *Ordinate* is in correlation coefficient units multiplied by one hundred. For explanation see test.

identifies the code numbers of the most closely related species while line 2 gives the value of the correlation coefficient of the relative with the isolated species concerned. Line 3 shows the level of the base line for the group of relatives up to and including the species listed in line 1. Thus in the case of species 4 the base line for species 2 and 6, its closest relatives, is .85 and remains at that level upon addition of the next ten relatives (species 8, 14, 15, . . . 7).

Figures 10 and 11 show the levels of the base lines of table 4 in graphic form. On careful study the lines fall into a number of more or less definite types. Type I, as exemplified by species 1, 31 and 97 and group 43–44, includes species whose nearest relatives are themselves a closely knit group, while more distant relatives (among the 12 studied) are still fairly highly related. Species 1, for example, is clearly closer to the 2–16 group (of fig. 5) than it is to group 17–18. There is,

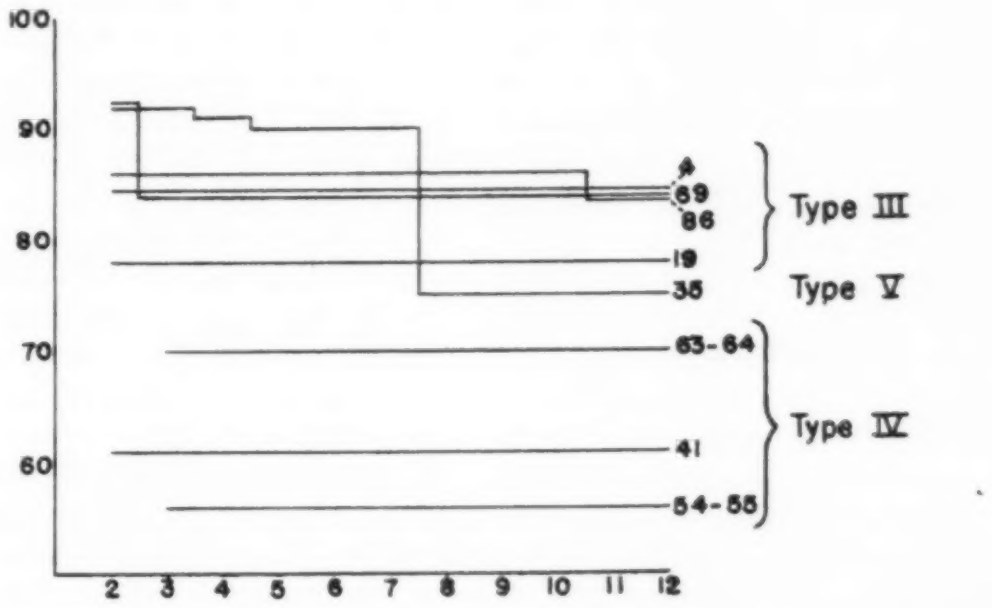

FIG. 11. Base lines of relatives of isolated species types III and IV and of a non-isolated species, type V (data from table 4).
Abscissa and *ordinate* as in figure 10. For explanation see text.

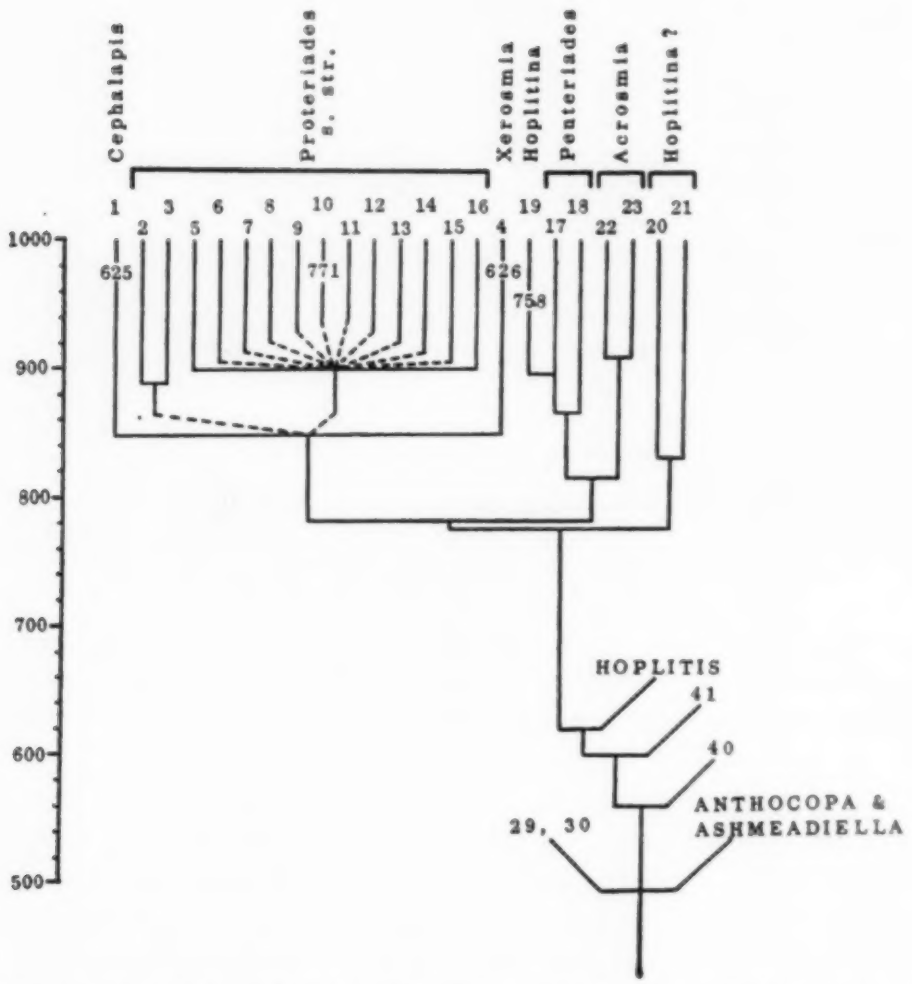

FIG. 12. Diagram of relationships for the genus *Proteriades* modified from figure 5 after consideration of the pregroup-exgroup problem.

Isolated species 1, 4, etc., shown as pregroup species in figure 5, are now shown as exgroup derivatives of various stems. Numbers inserted in the lines leading to these species indicate the levels, in terms of the scale at the left, at which the lines for these species joined their relatives in figure 5. These numbers therefore give an indication of the unusually large amount of specialization that has occurred in these species.

however, no evidence linking it to either 2–3 or 5–16 in preference to the other group. While the previous study had shown that 2–16 and 17–18 were much closer to each other than they were to 1, and consequently species 1 had been depicted as isolated, the present findings indicate that 1 has closer affinities to the 2–16 stem than to the 17–18 stem. This suggests that species 1 is a greatly modified offshoot from the 2–16 stem. In relation to species 1 the pregroup-exgroup terminology is not too appropriate. Species 1 is pregroup to groups 2–3 and 5–16, but exgroup to 2–16. As long as the correct relations are depicted, however, the terminology is secondary; moreover all intermediates must exist between pregroup and exgroup species. Reasoning similar to that employed in the case of species 1 leads us to regard species 31 as a modified offshoot of the 25–27 stem, species 97 as derived from stem 88–94, and group 43–44 as derived from stem 47–48, possibly from species 42.

In type II, including species 40 and 50 and perhaps species 58, as well as group 29–30, the near relatives of the isolated species are not too closely related while the more distant relatives are equally distant from each other. The isolated species concerned would more likely be pregroup species to their closer relatives. Species 40 appears closest to stem 25–28, while species 50 appears to have been placed correctly by the variable group method. Species 58 apparently branched from the 51–52 stem. Group 29–30 may

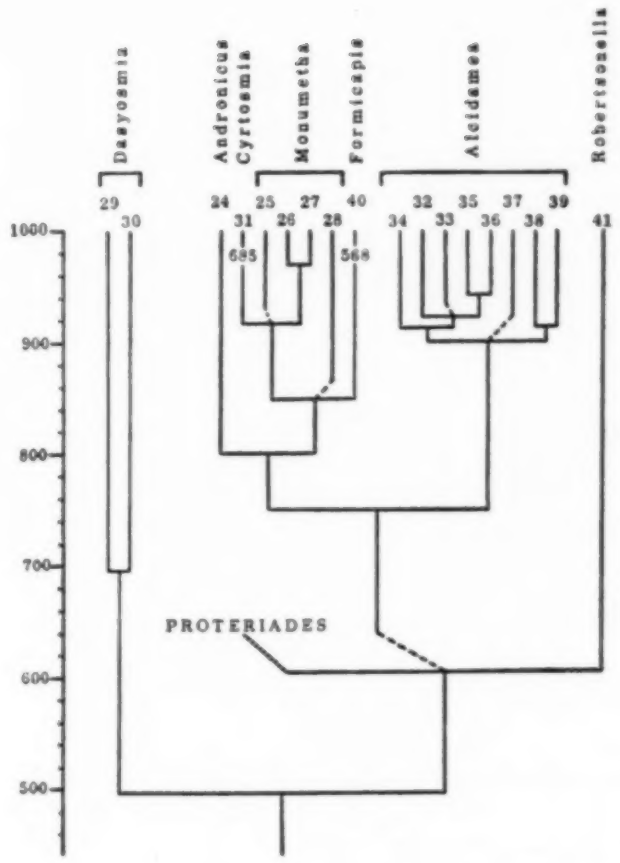

FIG. 13. Diagram of relationships for the genus *Hoplitis* modified from figure 6 after consideration of the pregroup-exgroup problem. (*Anthocopa* and *Ashmeadiella* stem omitted by error at level 500.)

Explanatory comments as in figure 12.

be a much altered branch off the 24–39 stem.

Type III includes species 4, 19, 69 and 86. Each of these seems equally and fairly highly related to a large number of species indicating that they originated from a large species group. Species 4 appears equally closely related to 2–3 and to 5–16 and is therefore represented as originating from the 2–16 stem. Species 19 has close relations to the 5–16 and 17–18 groups but curiously not to 2–3. Since it has a higher correlation coefficient with species 17 than with any other species it might be suggested that it originated from an ancestor of that species. Species 69 appears closest to 67–72, while 86 shows relations with 67–84.

We have designated as type IV species such as 41 or groups such as 54–55 and 63–64, which do not show close relations to any species. Those species to which they are somewhat related are not in a group by themselves but are scattered throughout the study. Such evidence would indicate that species of this type are isolated pregroup species. Their positions on the diagrams of relationships, therefore, have not been changed.

Finally species 35 was chosen as an example of a non-isolated species, type V. An analysis similar to the ones carried out above corroborates the position assigned by the variable group method.

New diagrams of relationships were constructed (figs. 12–15) based on a composite view of morphological resemblance as shown by the variable group method and relationship by descent as found by the above procedure. These diagrams are reasonably close to the original figures 1 and 2, indicating a surprising amount of agreement between the subjective and the more objective methods.

It should again be emphasized that the variable group method cannot by itself supply a diagram of relationships by descent. It can indicate static, nonhistorical relationships only. However, when another technique is applied in conjunction with the variable group method a diagram of relationships which is phylogenetically meaningful can be obtained.

Classificatory Problems

A difficulty arising in any large systematic study is the question of assigning hierarchic rank to taxonomic categories. A systematist has two purposes in mind when establishing a hierarchy of categories: (1) He would like to set up a so-called natural system of relationships which would indicate *both* common descent as well as degree of diversification (or amount of evolutionary change subsequent to the divergence of phylogenetic lines); (2) he uses categories as a convenient means of organizing his information (i.e., the many different species with which he has to deal). It is regrettable but true that the present system of nomenclature cannot easily meet both purposes. Moreover even in purpose (1)

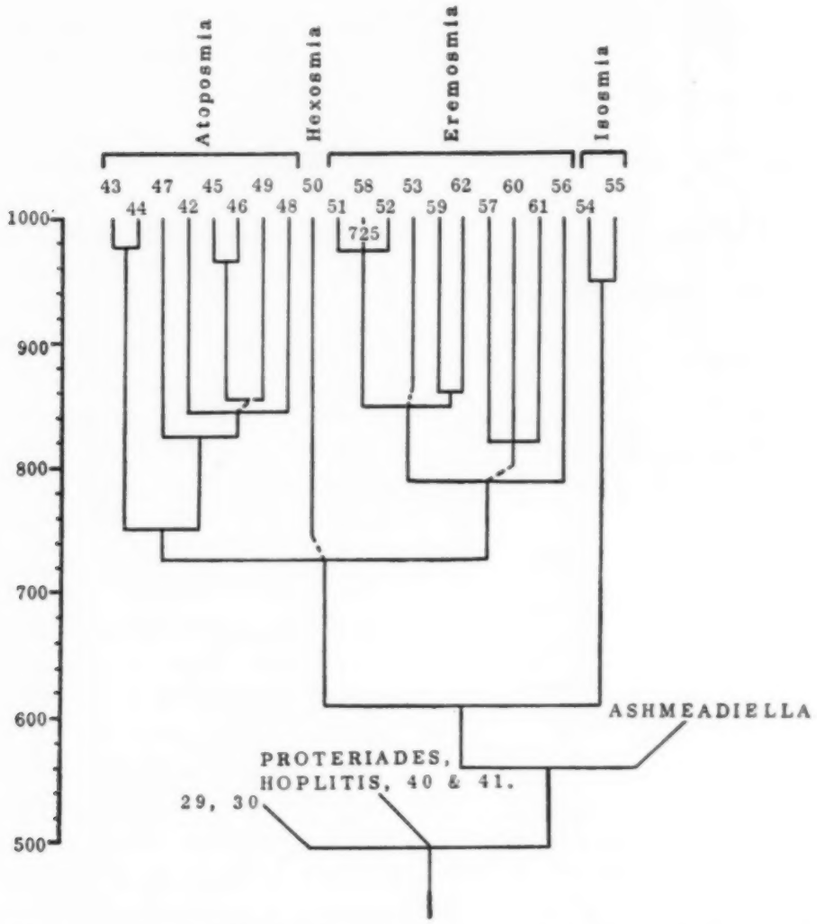

FIG. 14. Diagram of relationships for the genus *Anthocopa*, modified from figure 7 after consideration of the pregroup-exgroup problem.
Explanatory comments as in figure 12.

the present nomenclatorial system cannot always show both common descent and evolutionary divergence. Thus, referring to figure 9, it will be found difficult to devise a nomenclature to differentiate situations A and B. If we call both A and B genera, for example, then we reveal nothing of the different origin of X in the two diagrams. On the other hand calling 1–4 and X separate genera in A but the same genus in B implies that X is closer to 1–4 in B than it is in A, which is true from the viewpoint of time of divergence, but not from that of evolutionary change.

The two authors have divergent views on this issue. C. D. M. would prefer a more or less subjective approach, utilizing information gained by the statistical method but modifying it with knowledge of probable actual phylogenies obtained in a variety of ways. He considers the convenience of a classificatory scheme important. For example, he would prefer for the sake of convenience, that the size of groups recognized be more or less inversely proportional to the degree of difference between them. He is joined in these views by many present day systematists whose position might be summarized in the words of Mayr, Linsley and Usinger (1953, p. 48): "A genus is a systematic category including one species or a group of species of presumably common phylogenetic origin, which is separated from other similar units by a decided gap. It is suggested for practical reasons that the size of the gap be in inverse ratio to the size of the unit. The latter qualification will prevent the recognition of unjustified monotypic genera." Thus while *Proteriades* (species 1–24) was labeled a genus by C. D. M.,

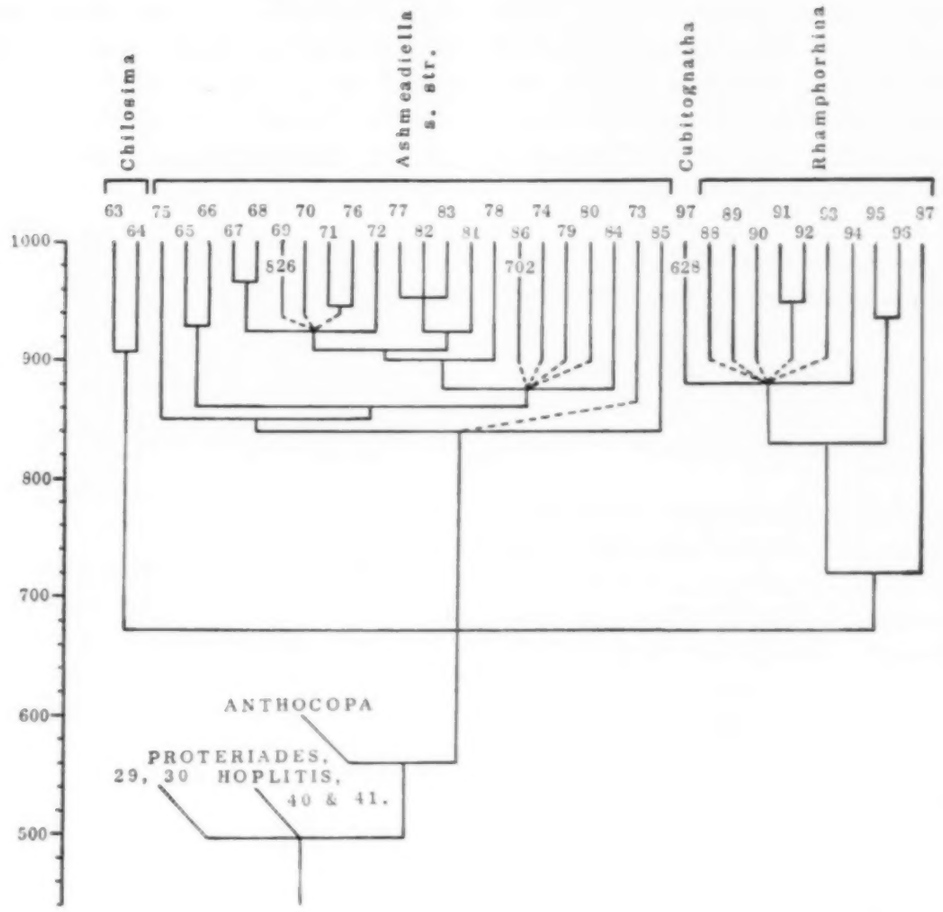

FIG. 15. Diagram of relationships for the genus *Ashmeadiella*, modified from figure 8 after consideration of the pregroup-exgroup problem.
Explanatory comments as in figure 12.

Dasyosmia (species 29–30), which is equally different from the other genera, was not accorded generic rank.

On the other hand, R. R. S. would like to see uniform and objective standards applied to the recognition of categories. Not being a practicing taxonomist he would prefer that the classification reflect the quantified relationships rather than considerations of convenience. R. R. S. feels that the only method that even approximates a realistic description of relationships is a pictorial one and that the hierarchy of categories can try to indicate either descent or amount of divergence (or relationship) between stems, but not both. He maintains that in the example cited above the nomenclature reflects descent in one case (by including *Dasyosmia* in the genus *Hoplitis*) but indicates divergence in another case (calling *Proteriades*, which also descended from *Hoplitis*, a separate genus). Rather than use categories in this ambiguous manner he would prefer to utilize them to show divergence or relationships only, leaving the description of descent to diagrammatic representation. He would therefore urge both "lumping" and "splitting" wherever indicated by the variable group analysis, i.e., he has no *a priori* objections to either monotypic or inordinately large genera or subgenera.

Proceeding on the above line of reasoning R. R. S. drew two arbitrary horizontal lines across figures 5 to 8. The levels of these lines, which are the same for all four figures, were determined by having them cut the ordinate at a point which would result in the number of categories (i.e., genera and subgenera) closest to the number established by

C. D. M. prior to the present study. The upper broken line might thus be called the subgeneric line and the lower, the generic line. By this method he would establish five genera: a joined *Proteriades-Hoplitis, Dasyosmia, Formicapis, Anthocopa* and *Ashmeadiella.* A subgeneric classification based on the upper line would include more monotypic subgenera than before (about half of them) as well as one newly constituted very large subgenus resulting from joining of most of (old) *Proteriades* with *Acrosmia* and *Hoplitina.*

C. D. M. as the advocate of the subjective approach, however, maintains that size of gaps as well as degree of difference is important and variable. He finds complications such as in the first matrix where species 54 and 55 had higher correlations with species 80 (in *Ashmeadiella*) than with any other species of *Anthocopa.* Obviously the intergeneric gap here is small. He would therefore prefer the flexibility of the subjective approach which permits consideration of degree of difference, size of gaps between groups, number of species in the groups, etc.

Taxonomic Consequences

The taxonomic consequences of the application of the objective approach can be determined by anyone who wishes to study figures 5 to 8 and 12 to 15. The classification proposed below is based on an admittedly subjective interpretation, but it makes extensive use of results of the objective statistical work summarized in figures 12–15.

In a number of cases a final decision was delayed because of excluded species which might shed light on certain dubious relationships. This classification should be examined in relation to table 1.

1. Genus *Proteriades.* Retained as a genus and expanded to include *Hoplitina* and *Acrosmia.* Can be distinguished from *Hoplitis* in its new, more restricted sense by the apical process, bearing specialized hairs, on the sixth metasomal sternum of the male; near absence of an apical flange on the sixth tergum of the female; broader and shallower basal sulcus on the first tergum than in *Hoplitis,* etc.

a. Subgenus *Cephalapis:* Unchanged. Seems to be an exgroup species from *Proteriades* s. str. but is sufficiently different to merit a name.

b. Subgenus *Proteriades* s. str.: Expanded to include group B of subgenus *Xerosmia* (table 1). Species 10 is merely a specialized "exgroup" member of this group.

c. Subgenus *Xerosmia:* Includes species 4 (group A) only, which has been amply demonstrated to be very different from the 5–16 (B) group. It is probably an exgroup species from *Proteriades* s. str. but is so different as to merit a name.

d. Subgenus *Penteriades* new subgenus. Type species: *Osmia remotula* Cockerell. Includes species 17 and 18 of *Proteriades.* Can be distinguished from other *Proteriades* by the five-segmented maxillary palpi, the absence or near absence of hooked or wavy hairs on the galeae and labial palpi of the males, and the bulging upper part of the clypeus of the females.

These bees are doubless similar to the primitive stock from which species 1 to 16 arose. They are also similar to *Acrosmia,* whose species evolved along quite different lines.

e. Subgenus *Acrosmia:* Unchanged in content.

f. Subgenus *Hoplitina:* Unchanged in content.

From figures 5 and 12 it appears that this group should be divided. We are inclined to take this action, but have delayed until we obtain both sexes of species known from only one sex, which were excluded from our study and which may be important in this decision.

2. Genus *Hoplitis*. *Hoplitina* and *Acrosmia* removed to *Proteriades*.

The species 41, 40 and 29 and 30 are quite atypical. However, the relations of *Hoplitis* to *Anthocopa* should be further investigated in Eurasia before a decision on the status of these species is reached. At the moment it appears most useful to recognize *Hoplitis* as a large primitive group from which *Anthocopa* arose at a rather low level on the diagram, *Proteriades* at a higher level. Subgenera of *Hoplitis* remain unchanged in content.

3. Genus *Anthocopa*. Unchanged in content.

a. Subgenus *Atoposmia:* Unchanged. Species 47 is probably an exgroup member of group 42–48.
b. Subgenus *Hexosmia:* Unchanged.
c. Subgenus *Eremosmia:* Species 54 and 55 excluded. Species 58, probably an exgroup member, is included in *Eremosmia*.
d. Subgenus *Isosmia*, new subgenus: Type species: *Anthocopa rubrella* Michener. Includes species, 54 and 55; can be distinguished from other *Anthocopa* by having the form and appearance of *Ashmeadiella;* second to fourth metasomal sterna of male subequal in size; apical margin of sixth metasomal tergum of female in the form of an unusually broad flange.

4. Genus *Ashmeadiella*. Unchanged in content.

a. Subgenus *Chilosmia:* Unchanged.
b. Subgenus *Ashmeadiella: Titusella* placed as synonym by C. D. M. since 86 appears to be an exgroup representative of 67–73. Species 69 is doubtless an exgroup form, phylogenetically related to the large central group of the subgenus (see fig. 15).
c. Subgenus *Titusella:* Merged with *Ashmeadiella*.
d. Subgenus *Rhamphorhina:* Subgenus *Corythochila* placed as synonym. Species 87 is included for the present. It is isolated and has primitive features (a pregroup species) but is connected to 88–94 by a few species known only as females and hence excluded from our study.
e. Subgenus *Cubitognatha:* Unchanged because of very distinctive features although 97 is an exgroup representative of 88–94.

Applicability and Objections

Applicability of Method

A number of implications and applications of our procedure as well as some objections to it have been discussed in previous sections. It remains for us to enter upon the general question of applicability of the method and to discuss several objections which we have so far not had occasion to mention.

The method described above provides a means whereby relative similarities among species of any group may be determined on an objective basis provided numerous characters differentiating the species are known or can be discovered. The relative similarities can then be used (with or without subjective interpretation) to form a classification which in our case resembled that devised previously by classical systematic methods. This similarity might be used by an advocate of either method to support his favorite technique.

For many groups already studied by competent systematists, our procedure will corroborate and quantify relations already well-known. It is satisfying to the systematist to have his findings substantiated in such a manner and at the same time our method can serve as a check against erroneous conclusions. Taxonomy, more than most other sciences, is affected by subjective opinions of its practitioners. Except for the judgment of his colleagues there is virtually no defense against the poor taxonomist. Above the species level there are no experiments that can be repeated and shown

invalid, no mathematical or symbolic reasoning that can be demonstrated to be in error. It is here that our method has advantages since it will give results disregarding subjective opinions if only characters are chosen properly (see below) and computations performed correctly. Our method may be no better than the work of the best systematist, although it provides much information useful to him; but it is far better than the work of the poor one. (A few years ago C. D. M. placed species 4 in *Anthocopa*. The error was detected and corrected by standard methods; it would have been detected by the statistical method also.)

Serological or chromatographic studies may perform a similar function if the serum proteins are sufficiently representative of the genotype of the animal; more likely they merely represent some additional characters to be evaluated along with the rest.

Presumably our method could be applied to almost any group of organisms, although among lower categories in plants, where reticulate evolution has occurred, the interpretations might differ. Even in groups having extensive fossil records, our method could provide a quantitative measure of amount of difference, hence of evolution, between various types. It could not be applied to groups exhibiting few characters, but we believe that all groups, except possibly some microorganisms, have enough characters if students will undertake to find them, although in some groups the characters will be largely internal or for some other reason more difficult to find than in bees.

Presumably any of the characters could be used, whether morphological, physiological, or behavioral. When using morphological characters we would recommend choosing characters from all parts of the body and would suggest that any duplicate characters (same character for different sexes, growth stages, or on serially homologous organs) be included as single characters. Characters that are consistently and obviously physiologically coupled should be considered as a single character. Characters should never be organized in such a way that a particular state of one obviously requires a particular state of another. A maximum number of 9 states per character would simplify calculations and should provide an adequate number of classes for most characters.

The number of characters used in such a study as ours should not be less than sixty, as the confidence limits for the correlation coefficients become too large with fewer characters. Also, the sampling of the genetic spectrum might become too incomplete with fewer characters. Since confidence intervals decrease slowly and genetic differences between species in a group are not inexhaustible, additional characters do not proportionally increase the value of such a study. However, little computational labor is added by more characters; if they can be found and recorded, they might as well be used.

As the method is independent of matrix size, it would work with fewer species than we used. We see no reason why it should not be used in a study of relationships among higher categories, such as genera and families. Among very high categories (classes, phyla) difficulty would be encountered, we suspect, in finding enough characters which are (1) known to be homologous among the various categories and (2) not so variable within some of the categories as to make coding of character states an impossibility.

As we have indicated already, we do not consider the method definitive; we hope to improve the method ourselves by factor analytic study and we hope to undertake similar or more refined studies. We do believe that it is a step in the right direction, that is, it decreases the subjective bias in systematic procedure.

As to its economic feasibility, we can point out that our total costs for assistants and IBM machine rental were about $300. Costs would decrease rapidly with fewer species and smaller matrices. Moreover, high speed electronic computers are in-

creasingly widespread. Such machines could handle the entire computation, from raw data to completed correlation matrix, without human intervention in less than an hour, or about one-twohundredth of the time it took us to obtain the same information with punched card machines! Obviously the computational difficulties for such work are rapidly diminishing.

Objections to the Method

While the method here suggested provides an opportunity to quantify the degrees of relationship between various species or groups, the biological reliability of this quantification has yet to be examined. A consideration of certain species pairs gives some idea of the reliability of the information on levels of the forks found in figures 5 to 8. Species 67 and 68 are shown as correlated to a high degree (.96), while species 63 and 64 are less correlated (.91). A study of the actual differences shows that the principal reason for the lower correlation of the latter pair is that one of them has both the femora and the abdomen largely red, the other has these parts black. Thus in two characters, each having four states, these species fall at opposite extremes. Therefore their correlation coefficient was appreciably lowered. Probably the coloration of the femora and of the abdomen are causally correlated in 63 and 64, so that they ought to have been treated as a single character for this species pair, although this is obviously not so elsewhere in *Ashmeadiella* where forms with red femora and black abdomens, and with black femora and red abdomens, are well known. In any event, 63 and 64 are actually very similar and may not be specifically distinct. This shows that at the higher levels in the tree more characters are needed (whenever they can be found) to make the levels of forks more reliable. There were only five characters showing any difference in states between 63 and 64 and seven between 67 and 68. Also it shows that base levels as different as .91 and .96 may not be significant biologically even though they are statistically. These species pairs were selected for comment because the information regarding them in figure 8 seemed to C. D. M. contrary to his impressions as to relationships. Other similar pairs in our diagrams agreed better with his views. At lower levels in the tree we believe that smaller differences in correlation coefficients (levels of forks) are more significant biologically because of the much larger number of differentiating characters involved.

Another commentary on our method, if not an objection, at least suggests that the method is unnecessary. By working first at one classificatory level, then at another, the systematist is able to work with a large number of characters. He does this by ignoring, at each level, all of the characters that are constant within the group he is studying and also those that are variable within the subgroups. Thus if he is studying relationships in a species group, most of the characters are constant and he has only to consider the few that he can find varying among the species. If he is studying generic relations, he ignores characters constant within the subfamily and also, to a considerable degree, those variable within genera, considering only those varying among the genera. Obviously systematists can do good work by their method; we do believe, however, that a method considering the totality of relationships, using all characters, has advantages. In addition to those mentioned earlier, it makes possible consideration of significant trends among specific characters at generic and higher levels. Such trends are commonly ignored by systematists except for the statements that a certain genus *usually* exhibits a particular character.

We have no belief that our method will replace standard systematic procedure, although we believe it may frequently be useful to check such procedure by a statistical approach. The question arises, whether the statistical method could be applied to a systematically unworked or

little worked group. It is obvious that it could not be used until the species are accurately distinguishable, sexes properly associated (unless we are to base the systematics on only one sex), etc. Also, finding numerous characters is a major undertaking, even in groups like the bees which are relatively replete with characters. However, the method should have great advantages for groups containing numerous recognized species whose relationships are unstudied.

A final difficulty with the method concerns species exhibiting intraspecific variation for certain characters that have value in distinguishing species elsewhere in the group. In our study this was a minor problem. Only for a very few species which vary, for example, in the extent of red on the abdomen, was it necessary to record a character state code characteristic of the most widespread subspecies (or the commonest color form) and ignore variability among subspecies (or individuals). However, the problem is certain to be a major one, as already indicated, in any such study in which the objective is to determine relationships of higher categories without consideration of the included species.

Summary

The authors have developed a statistical method for describing relationships among species and higher systematic categories. The method is based on an analysis of a matrix of species-×-species correlation coefficients computed from a large number of characters for each species and has been called the weighted variable group method. Diagrams of relationships resembling phylogenetic trees are constructed and, on the assumption that morphological resemblance is with certain exceptions inversely related to amount of evolutionary divergence, phylogenetic relationships may be hypothesized.

The method is applied to a group of four genera of bees in the family Megachilidae. One hundred and twenty-two characters were studied for 97 species of this group. Relationships between the species in this study as visualized by Michener before the statistical analysis are compared with relationships as found in the analysis by Sokal. Good agreement between the two sets of data is found. The few exceptions to this rule are discussed. Some changes in the classification of these bees are made as a result of the analysis and two new subgenera are named. However, the paper serves primarily to illustrate a method that can be used to remove some of the subjective bias from taxonomy.

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SEX CHROMOSOMES IN AMNIOTA

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INTRODUCTION

During the past 25 years the chromosomal conditions in Vertebrates have been the subject of a great many investigations. With some justification it has been hoped that such work might add to our present knowledge of the evolutionary relation between the different classes of Vertebrates and, in this connection, special attention has been given to the sex chromosomes. Unfortunately the findings of various cytologists are by no means in agreement and the present note represents an effort to reach a final conclusion, at least so far as the Amniota are concerned. The case of the Anamnia is much clearer: with the only exception of the fish *Mogrunda obscura* where Nogusa (1955) has described in the male a heteromorphic bivalent of the X-Y type, we may assert that, as a rule, there is no cytological expression of the digamety in Fishes and Amphibians (Matthey, 1949).

Application of a modification of Makino and Nishimura's (1952) squash technique to all major groups of the *Amniota,* except for the *Rhynchocephalia,* has led us to the following conclusions:

REPTILES

Lacertilia. From 1934 to 1956 two opposite opinions have been held: the Japanese school (Oguma, 1934; Makino and Asana, 1948) claims that in *Lacerta vivipara, Calotes versicolor* and *Sitana ponticeriana* the female has one chromosome less than the male (♀ZO, ♂ZZ). The Swiss school (Matthey, 1943; Margot, 1946) does not find any difference between the chromosome sets of the two sexes and concludes that there are no morphologically recognizable heterochromosomes in *Chamaeleon vulgaris, Anguis fragilis,* and *Lacerta vivipara.* The validity of this conclusion was further proved for *Chamaeleon bitaeniatus, C. dilepis, Brookesia stumpffi,* and *Lacerta vivipara* in two recent papers by Matthey and van Brink (1956) by using the above mentioned squash technique instead of the paraffin method which was used for the former studies.

Ophidia. We examined 23 embryos of one pregnant female of *Natrix rhombifera.* Every embryo contained a large number of mitoses, all of which showed 36 chromosomes without exception (fig. 1). No morphological differences between the chromosome sets of different individuals could be observed. Since it is highly improbable that all the embryos would have been of the same sex, the conclusion drawn in the case of the lizards may be extended to the snakes.

Crocodilia. In a young female of *Caiman sclerops* 42 chromosomes were observed (fig. 2) and the same number was found in the male (fig. 3). Here again there is no difference in number or morphology between the two sexes. In passing, it should be noted that *Alligator mississippiensis* (Matthey, 1947). has 32 chromosomes, ten of which are large metacentrics; as the latter chromosome type is missing in *Caiman sclerops,* a beautiful illustration of Robertson's rule is shown by these two species.

FIG. 1. *Natrix rhombifera,* embryo.
FIG. 2. *Caiman sclerops,* female
FIG. 3. *Caiman sclerops,* male.
FIG. 4. *Emys orbicularis,* female.
FIG. 5. *Tachyglossus aculeatus,* male.

Chelonia. For this group the Japanese school (Oguma, 1937; Nakamura, 1949; Makino, 1952), studying turtles of the genera *Amyda, Caretta,* and *Chelonia,* adopted the formula Female = ZO, Male = ZZ. Our own observations on *Emys orbicularis* permit the conclusion that the diploid number in the female is even (2N = 50) (fig. 4). In the same species, Matthey (1931) likewise counted 50 chromosomes in the male. On the basis of our observations we are absolutely convinced that for counting purposes our squash technique is far superior to the sectioning technique and that it gives preparations which can be analyzed without any subjective errors.

BIRDS

Investigations of the Japanese school (Susuki, 1930; Oguma, 1938; Yamashina from 1944 onwards) have led to the identification of an odd chromosome in the female; this Z chromosome is generally represented by an element of medium size, in *Gallus* for example by the fifth largest chromosome. Apart from this, the Japanese authors claim that the total number of chromosomes is uneven in the female (Female = ZO; Male = ZZ). Matthey (1949), without denying the quality of the Japanese observations, expressed his doubt on this subject. Van Brink and Ubbels (1956) recently confirmed that the fifth largest chromosome in the female chicken is actually unpaired. On the other hand, the Dutch authors consider the exact determination of the total chromosome number (78 in *Gallus*) beyond the possibilities of cytological technique. Hence we may admit the existence of a morphologically expressed female heterogamety, whereas the question as to whether there is a ZO or ZW mechanism cannot yet be decided.

MAMMALS

Monotremata. The fragmentary observations of White (1945) on the spiny anteater and of Matthey (1949) on the duckbill showed the bird-like character of the chromosomal formula of this group of animals. According to Matthey's estimate, the diploid number of *Platypus* is 70 ± 10.

Examination of a male *Tachyglossus aculeatus* made it possible to fix the diploid number at 62 or 63, in spite of the fact that the spermatogenetic activity of the individual under study was low. Even though meiotic divisions were lacking in our material, the existence of male heterogamety is clearly proved by the presence of a large, unpaired acrocentric (fig. 5) that is superior by one-fourth of

its length to the members of the largest pair of autosomes. It follows that, as in other mammals, the male sex is heterogametic although the general chromosomal morphology of the *Monotremata* places this group near to certain *Reptiles* (*Chelonia*) and to the *Birds*.

Marsupialia and *Eutheria*. The formula Male = XY; Female = XX, is general. Certain cases of multiple sex chromosomes are known to exist in *Marsupialia* as well as *Eutheria*. In the *Microtinae* of the genus *Ellobius* (Matthey, 1954) both sexes possess 17 chromosomes, whereas this number of 17 has also been found in *Chilotus oregoni* it is therefore thought that in the latter case the digamety is of the XO type.

Summary

The cited observations put an end to a discussion that lasted for a quarter of a century. Morphologically distinguishable sex chromosomes are absent in *Reptilia*, as is known to be the case also for the *Anamnia;* the females of *Birds* and the males of *Mammals* are heterogametic.

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PHASE AND MOULTING POLYMORPHISM IN LOCUSTS

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Introduction

According to the Phase Theory formulated by Uvarov (1921, 1928), locusts exhibit a graduated type of density-dependent dimorphism. So marked is this dimorphism that the phases of most common locusts were once accorded specific rank. Phase polymorphism is characterized by two limiting forms linked by a whole range of intermediates; the form found in bands or swarms is known as phase *gregaria,* and the sedentary isolated form as phase *solitaria:* intermediate or *transiens* forms are designated by the terms *congregans* and *dissocians,* according to the direction of the phase-change.

The ideas presented in this essay stem from different kinds of morphometric analysis of the locusts. First, the occurrence of an extra moult in some individuals is found to be part of a twin homeostatic growth-regulating process during ontogeny. The polymorphism that ensues is stochastically related to the density of the parental population. This density-dependent quality, though inherited, is not genically determined. Careful study of the wing-pads of the developing nymphs distinguishes insects whose innately determined phase status is not in equilibrium with the density of the larval population of which they are part. *Congregans* and *dissocians* nymphs form two separate categories of this *transiens* phase, illustrating at once the dynamic conception of phase polymorphism and the existence of hysteresis in cycles of density-dependent changes of shape in locusts.

New methods for the quantitative study of form in locusts show that the generic differences between Red and Desert Locusts are independent of their phase polymorphism and of their normal growth. Finally, a necessarily speculative discussion of the role of polymorphism in the life-cycle of locusts is attempted.

Moulting Polymorphism

Recently revealed by investigations carried out on Red and Desert Locusts, one type of developmental polymorphism is characterized by differences of growth rate and of numbers of moults within a population.

Several hypotheses, propounded to account for the additional moult, have not brought a satisfactory solution. The assessment of the two types of development in morphological terms seemed profitable.

Red and Desert Locusts of various origins were reared and the head widths measured throughout the developmental cycle. Distinctions were found even at the eclosion of hatchlings. For Red Locust hatchlings, the head-widths of those eventually to have 7 instars ranged from 1.45 to 1.83 mm, those to have 6 instars ranged from 1.63 to 2.05 mm. With Desert Locust hatchlings, the 6 instar type began with headwidths ranging from 1.43–1.48 mm, and 1.47–2.25 mm was the range of the 5 instar type. In the same way, Remaudière (1954) found that only the smallest third instar female *Locusta migratoria migratoroides* pass through an additional moult, the larger females at this stage completing their development with one fewer moults.

The two distributions relating to the

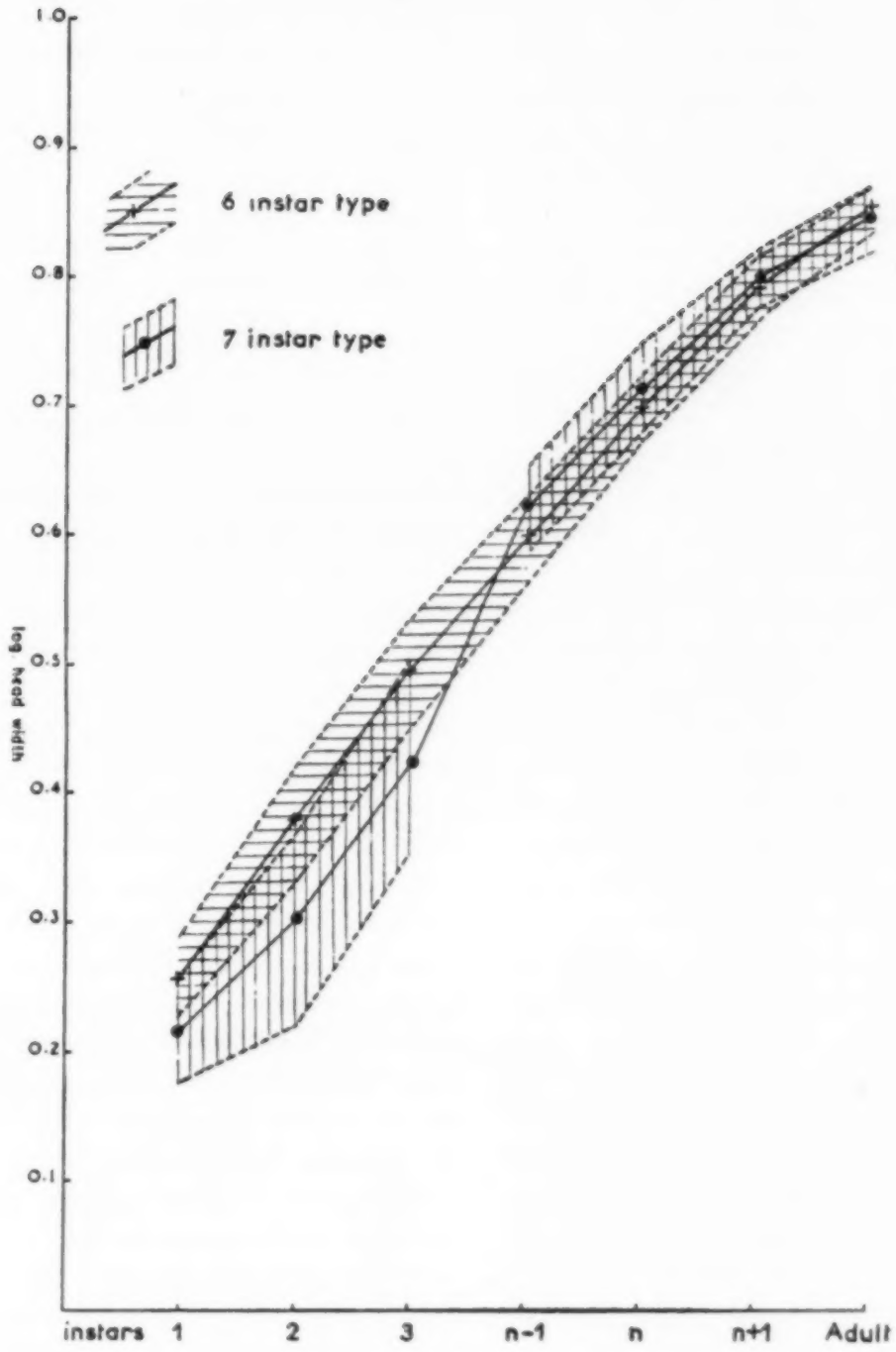

FIG. 1. Growth rates of male Red Locusts of each developmental type.

Red Locust are of roughly equal range, and overlap by about a third of the total range of head widths for the species. The developmental type of Desert Locusts with 6 instars, corresponding to that of the Red Locust with 7 instars, exhibits at eclosion a distribution of head widths, narrowly confined to the smallest values, which overlaps less with that of the 5-instar Desert Locusts than do the two Red Locust distributions with each other. For insects from any one egg-pod, the head width and weight are proportional, within the ranges studied. The constant of proportionality changes slightly, but significantly, from egg-pod to egg-pod, as might be expected from such a strongly allometric relationship.

No single egg-pod of the Red Locust was ever observed to yield hatchlings solely of one developmental type, whereas the majority of Desert Locust egg-pods yielded hatchlings of the 5-instar developmental type. The range of head widths of hatchling Red Locusts from a single egg-pod (about 0.25 mm) is wide enough consistently to cover a third or more of the total range: the two developmental

types should both occur, though in varying proportions, from most individual Red Locust pods since the range of head widths must normally cover at least part of the overlap region. For the Desert Locust the range of hatchling head widths within an egg-pod is about 0.35 mm so that the two developmental types will occur from any single pod only when the pod gives rise to hatchlings whose head width lies at the extreme lower end of the scale. Further, the range in headwidth of hatchlings emerged from typical pods is such that no pod is ever likely to give rise solely to the 6-instar type.

The central thesis of this work is expressed in figure 1. The differential growth of the two developmental types is shown for Red Locust males reared in isolation; the head width of each developmental type at each instar is accompanied by the 95 per cent fiducial ranges. Heads of Red Locust hatchlings destined to pass through 7 larval instars are narrower than those of larvae embarking on a 6-instar developmental cycle. Further, the rate of growth of the 6-instar type as a group is higher than that of the 7-instar group, so that the initial disparity of size is increased at each moult until that preceding the reversion of the wing buds. Much of this discrepancy is removed, however, by the intercalation of the additional moult undergone by the 7-instar group.

There is a second type of growth regulation in these locusts. Throughout the developmental cycle, the initially larger hoppers within any one group grow more slowly than do the smaller hoppers of the same developmental type, so that within each type the uniformity of the group is steadily promoted (table 1). Fuller details are given by Albrecht (1955) in his appendix A.

This steady reduction of the heterogeneity of each developmental type of Red Locust is reinforced, immediately prior to the reversion of the wing buds, by the virtual elimination of the hitherto progressive inequality of size between the

TABLE 1. *Growth-rate of Red Locusts according to their size at birth*

Sex	Developmental type	Initial headwidth (mm)	Mean increment per instar (mm)
Females	6-instar	1.96–2.00	1.291
Females	6-instar	1.61–1.65	1.310
Females	7-instar	1.81–1.85	1.248
Females	7-instar	1.51–1.55	1.286
Males	6-instar	1.91–1.95	1.274
Males	6-instar	1.61–1.65	1.307
Males	7-instar	1.81–1.85	1.242
Males	7-instar	1.51–1.55	1.266

developmental types by the additional moult of the smaller group. There are thus two mechanisms which ensure that, although the hatchling locusts present a wide range of sizes, these differences are eventually reduced both within and between the developmental types to give adult locusts relatively more homogeneous than the earliest instars.

Both genetic and developmental bases have been sought for the difference in size at birth (Albrecht, 1955). Selection experiments demonstrated that any genic control of developmental type is scarcely detectable; the one positive outcome of such experiments was a practicable technique for uncovering the influence of density in the preceding generation. Differences of density at the parental level induced differences in the proportions of the two developmental types among the offspring. Two sets of egg-pods were taken from areas of high and low densities. Those from high densities gave hatchlings whose head-widths ranged from 1.67–1.99 mm. From low densities these headwidths ranged from 1.47–1.68 mm barely overlapping the other distribution. The consequences of this difference in terms of developmental polymorphism have already been noted.

Further experiments confirmed the triple relationship between head width, the number of larval instars, and the integrated density of the parental population (Albrecht, 1955). The smallest hatchlings, obtained from females that have been in isolation practically all their lives, subsequently undergo the type of

life cycle with the extra instar. The density of larvae during the earliest instars preceding the intercalated moult does not influence the incidence of this supplementary moult.

Relation Between Eye-Stripes and Instar Number

Volkonsky (1937) has demonstrated that the number of eye-stripes is uniquely related to the number of moults undergone by the insects. The first instar larvae bears one eye-stripe, and a further stripe is added at each moult. This proposition was denied by Roonwal (1947) but further work (Albrecht, 1955) shows that there is a direct correspondence to which no exceptions have been found. Other characters which may be regarded as imprints of the moulting process are the number of antennal segments and the innervation of the true wings of locusts. Remaudière (1954) shows that the female migratory locusts which have passed through the supplementary instar possess antennae having 22 segments instead of 20 or 21, and at least seven axillary veins. Migratory locusts do not show eyestripes.

The Distinction Between Congregans and Dissocians Locusts

Further evidence is available from a study of the wing-buds in the course of their development. These organs are of special interest to field workers, being visible to the naked eye. Michelmore and Allan (1934) described Red Locusts passing through 6 larval instars. At that time a locust invasion of the East African Territories from which the locusts were taken was in progress, and these specimens were almost certainly the progeny of swarms. Burnett (1951) described the developmental cycle with 7 larval instars, this time from locusts whose parents were reared in comparative isolation. Figure 2 shows these two developmental types, and to simplify the description of the diagrams, the type with 7 larval instars is called the "Burnett" series and

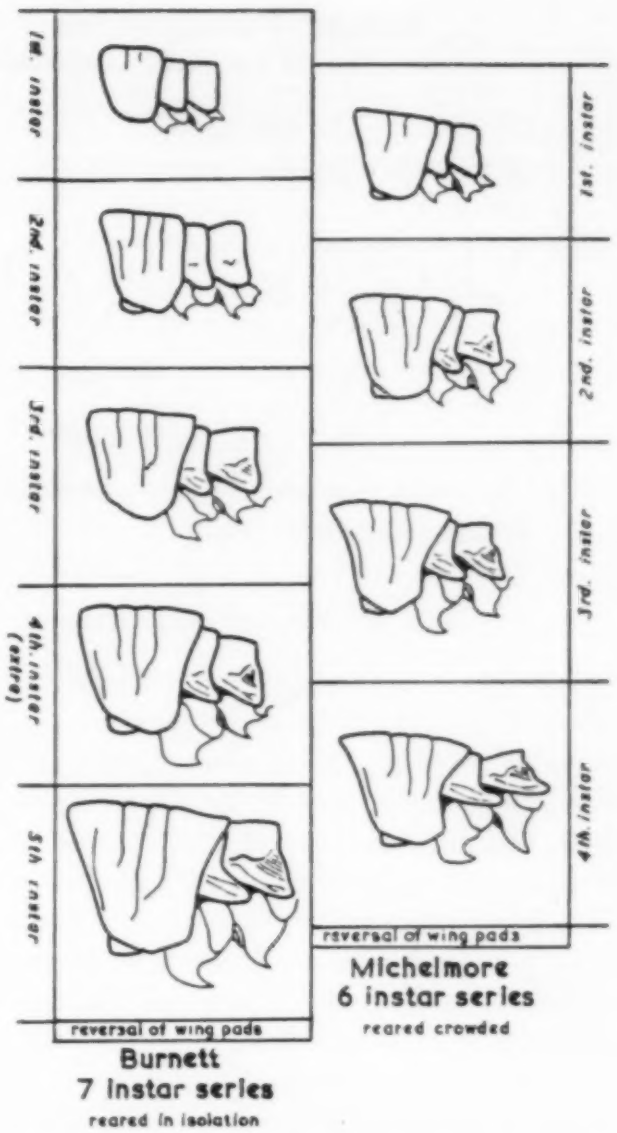

Fig. 2. Structural limits of wing pads of developmental types of Red Locust.

that with 6 such instars the "Michelmore" series.

Drawings of the wing-buds from the Burnett series faithfully represent insects passing through 7 larval instars when these are reared in isolation. Drawings of the Michelmore series accurately reflect the development of insects passing through 6 larval instars only when these are reared crowded. Attempts to rear either the Burnett series crowded or the Michelmore series in isolation yield forms intermediate between structural limits shown in figure 3, for the instar preceding wing reversion. The drawings on the extreme left and right show the structural limits. The other drawings show the results of attempts to rear either series at the density appropriate to the

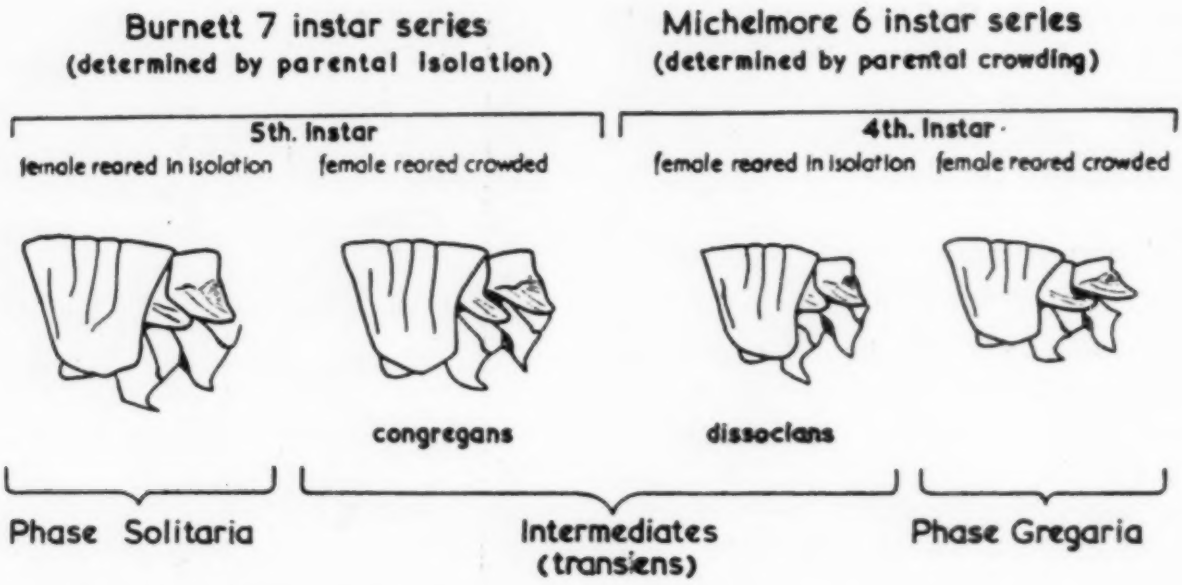

FIG. 3. The structural limits of the wing-pads during phase transitions.

other: the wing-pads of the Burnett series have an elongate and narrow aspect when the insects are reared at high density, thus tending to resemble those of the Michelmore series reared at high density. The wing-pads of the Michelmore series reared in isolation are wide and rounded and tend to resemble those of the other series reared in isolation.

In fact, each developmental type has a range of structural forms, and the discontinuity of form between them suggests that neither has sufficient plasticity fully to adjust itself to densities appropriate to the alternative.

Insects closely approaching one or other limiting form can only be obtained when crowding or isolation at the parental level is maintained during the lifetime of the progeny. Disparate densities of parent and progeny give rise to intermediate or *transiens* forms. Thus, figure 3 shows the Burnett 7-instar form, determined by isolation at the parental level. This form is typical of phase *solitaria,* when breeding is maintained in isolation, and may be considered as *congregans,* when the first increases in density are felt by the insect. Similarly, *dissocians* arises from the Michelmore 6-instar form reared in isolation, i.e., from the isolation of hoppers from crowded parents. Only the Michelmore form morphologically represents the typical *gregaria* phase, which requires the crowding at least two successive generations.

In any single population of common origin, if *solitaria* insects are accompanied by intermediates, these will be predominantly of the *dissocians* type, whereas if *gregaria* insects are accompanied by intermediates, these will be predominantly of the *congregans* type.

Samples of hoppers collected in the Rukwa valley (Katisunga, Tanganyika Territory) in 1952, were obtained from a dense concentration of hoppers. According to eye-stripe numbers, 81 per cent of the males and 78 per cent of the females belonged to the Burnett 7-instar form, indicating that the populations were dispersed in the preceding year, a fact previously known from scouting reports. The study of the wing-buds revealed the morphological phase status of the population. 81 per cent of the males, 78 per cent of the females, were *congregans* in the sense of figure 3, the remainder of the population being morphologically typical of phase *gregaria.* No *solitaria* or *dissocians* forms were present. In the Niombe district of the Central Rukwa in 1952, a sample was collected of 4th and 5th instar hoppers. Seventy eight per cent of the males and 58 per cent of the females belonged to the Michelmore se-

ries, a reflection of the high infestation in 1951. Only *dissocians* and *solitaria* were present, a few embarking on the transition *solitaria* to *congregans;* no *gregaria* were present.

This reasoning applied not only to wing-buds, but also to other morphological characters, such as the female genitalia. However, genitalia react less than wing-buds to density, and the plasticity of the genitalia no longer masks the distinctions between the instars.

Density dependence of morphological structures is integrated throughout the development of the hoppers, and fixed in the adult. The relative discriminatory powers of the different characters can now be discussed.

The Quantitative Analysis of Form in Locusts

The study of changes of form in animals has made progress mainly where quantitative expression of such changes is practicable. The comparison of allometric indices has until recently been limited by the restriction that only two characters could be compared at once. This limitation has been lifted by the introduction of principle component analysis or of factor analysis, based on several characters (Burt and Banks, 1947; Cousin, 1948; Teissier, 1955).

Mahalanobis, Majumdar and Rao (1949) provide two related methods of multivariate analysis by means of which the differences of form among Indian castes and tribes can be reduced to comparisons along a few qualitatively distinct dimensions. A recent anthropometric contribution in the same vein is that of Mukherjee, Rao and Trevor (1955). The mathematical bases for these methods have been developed by Rao (1952) who has called the subject discriminatory topology.

In one of these methods, which has been used here, groups of locusts are distinguished as clearly as possible by combining their measured characters into a linear discriminant function. In these functions the characters are added together in such proportions that they afford the greatest possible separation between the groups of locust, which are, in effect, being pushed apart, by the addition of each new character, in a space of as many dimensions as there are characters (Fisher, 1938). This greatest separation between the groups, taking into account the correlations between the characters, is known as the generalized distance (D). If the coefficients in the discriminant function are $\beta_1 \ldots \beta_p$, and the differences between the mean values of the p characters for the pair of groups being considered are $d_1 \ldots d_p$, then

$$D^2 = \beta_1 d_1 + \beta_2 d_2 + \ldots \beta_p d_p$$

The quantity D is a pure number, and for the first pair of a series of groups D may be plotted on any scale. The generalized distances between each of the first pair of groups and the remainder may then be used to construct a chart. If the groups differ in but one fundamental dimension, the chart will collapse to a set of points scattered about a straight line.

If two fundamental dimensions of variation are concerned, the generalized distance chart occupies a plane surface. For three or more basic sources of variation, the chart becomes a solid model, although we are usually interested in the few most important dimensions. Such methods generalize the analysis of trends in variation which Anderson (1951) has used to determine hybridization between plant species.

Laboratory Desert Locusts reared in isolation differ from those reared crowded along a dimension distinctly different from that which contains comparisons between the sexes. By including the weight of the adults at eclosion as one of the characters, and comparing the coefficients of this character when phases or sexes were being compared the comparison between the sexes was shown to be essentially one of size differences. This con-

Desert Locust ♀♀ crowded
Desert Locust ♀♀ isolated
Red Locust, ♀♀ crowded
Red Locust ♀♀ isolated
Red Locust ♂♂ crowded
Red Locust ♂♂ isolated

FIG. 4. Inter- and intraspecific differences in locusts.

clusion was verified for the Red Locust (fig. 4).

Cousin (1948) used quantitative morphometric methods to distinguish interspecific differences of form. Her suggestion that an hierarchy of structural types might be constructed to run parallel with, and complementary to, the dichotomous taxonomic classifications is particularly suited to the lower taxonomic ranks, including sub-specific levels, whose taxonomy is least firmly based on phylogenetic foundations.

We might be able to find out whether any recent speciation has sprung from an exaggeration of sub-specific differences, of a kind still recognisable in related species. Orthoptera, difficult to distinguish in a dichotomous key, are already being separated by the use of discriminant functions (Bigelow and Riemer, 1954). For the first time, we can study the form of insects quantitatively. Previously, Richards and Kavannagh (1945) noted that the only method by which several characters could be assessed together was the coordinate transformation method of D'Arcy Thompson. This method, they remarked, is essentially descriptive. Attempts to make it quantitative have not been conspicuously successful.

Representation of Interspecific Differences of Form in a Chart of Several Dimensions

In figure 4 discriminatory topology illustrates the relations between the inter- and intra-specific differences of form in locusts. Red Locusts from two populations from the Rukwa were used to provide measurements of the elytron and posterior femoral lengths and of the head-width. The insects from a highly dispersed population were estimated as having a density of 1 locust to 85 sq. yards. Another sample was too dense to permit of accurate estimates of density, though not quite as dense as some swarms of this insect. These insects were collected and measured by Dr. R. F. Chapman, of the International Red Locust Control Service, to whom we are greatly indebted. We are equally grateful to Dr. M. J. Norris for the same measurements on laboratory samples of the Desert Locust, reared by her at the Anti-Locust Re-

search Centre, London. Gunn, Hunter-Jones and Davies (1952) have found that the full morphometric expression of density differences is incompletely realized by insects reared in the laboratory so that the density-differentiated groups are described on the chart as relatively more and relatively less solitary.

The main conclusions from figure 4 are:

(1) Differences of form between the species are qualitatively distinct from intraspecific differences. This observation lends no support to the suggestion that the evolution of the two genera has involved the exaggeration of differences of size or phase already to be found within the structural type of the generic ancestors.

(2) The dimension of phase, reflecting differences of density, and the size dimension, which incorporates the sexual dimorphism, are common to both species.

(3) A framework of reference is thus established, subject to confirmation with other species, by which lesser taxonomic differences may be distinguished from phase variation where the need arises. Several characters which are used as criteria of phase status are also sensitive to ecological and subspecific variation.

Discussion

When so pervasive an influence as parental density has escaped attention, the impression that capricious and highly variable results are likely in locust experiments is natural. Finer discrimination is possible once this source of variation is controlled.

The "centres of growth" (Huxley, 1932) may not all be of one kind in locusts. Growth of the head capsule is modified by parental density, and is probably innately determined. Uvarov and Thomas (1942) suggest that pronotal form, which, like that of the head-capsule is density-dependent, develops as a response to the differential muscular activity of the thorax in the two extreme phases. Although Duarte (1938) was unable to find differences in the head-widths of newly hatched Migratory Locusts of either phase, he measured the width at the base of the mandibles, not at the apeces of the genae. Ramchandra Rao (1942) has shown that in Desert Locusts, at least, Duarte's technique would not disclose differences which are known to exist.

As Kennedy (1956) has emphasized, the theory of polymorphism which was the basis of early studies of phase in locusts has at times stopped short at the anatomical level of explanation, not considering such changes as a response to modifications of behaviour consequent upon changes of population density and other ecological factors.

It may be that this sequence of attitudes to locust polymorphism, whose history Kennedy unfolds, will recur in other fields. For instance, there are three supposedly distinct genera of Ephemeroptera whose larvae are quite unlike one another, yet the imagos are so alike that it is a moot point whether they can be distinguished one from another (Verrier, 1956). *Ephemerella* and *Torleya* occupy distinct habitats, and *Torleya* larvae have a more precocious developmental sequence. It now transpires that, together with the little-known genus *Chitonophora*, *Torleya* may constitute the generation of *Ephemerella* which matures precociously in the spring. These putative "genera" were erected on the basis of the marked larval disparities without knowledge of adult form. As Kennedy notes, even generic status has been given to groups of locusts on account of distinctions which ultimately proved to be density-dependent. The larval differences among the *Ephemerella-Torleya-Chitonophora* complex, although altogether more striking than those among larval polymorphs of locusts, may prove to be somewhat similar. Al-

though the different larval environments play the part of mutual stimulation between locust hoppers the idea of a common imago resulting from widely polymorphic larvae is basic to each train of thought.

The results outlined in this essay invite comparison with the phenomenon called "teleogenetic paragenesis" by Dalcq (1941). In his example, individual chicks with differing genetic constitution developed along different lines, yet ultimately converged to give similar phenotypes. Locusts have no known genic differences sufficient to establish the eyestripe polymorphs as different genotypes, yet two distinct developmental cycles lead to a common phenotypic expression.

As Wigglesworth (1954) notes, these courses of growth may be controlled by genes even though no such control has yet been demonstrated. No doubt the capacity for polymorphic development is latent in the genotype though its realization is effected by environmental conditions. Probably the course of polymorphism is determined by ovogenetic constituents which, differing quantitatively among the embryos, can simulate hormones in developing an all-or-nothing response. Of a similar nature, perhaps, is the differential rate of development of flour beetles, according to the parental diet (Reynolds, 1945).

Differentiation of developmental types commences before other than maternal influences can have been effective. The group stimulation of the female may lead to the preferential production of one polymorph in the progeny (Grassé and Noirot, 1947). Such a situation, in all probability, will profit the insects if there is a positive correlation between the conditions determining parental density and those existing during the immature stages of the next generation. Such serial correlations between adjacent periods are common. Among the factors which make up the weather, rainfall is likely to be important. Kraus (1955) has demonstrated such trends in tropical rainfall records, for instance in the Nile catchment area.

Red Locusts seem to have evolved twin growth-regulating mechanisms which remove the subsequent disadvantages of high initial variability.

Tentatively, our orientation is to regard moulting (eyestripe) polymorphism as an adaptive device by which an insect of fairly uniform adult dimensions is yet able to exploit a highly variable immature form hatched into the highly variable "mosaic" environment of many "Locust outbreak areas." It is early instars which suffer the highest mortality, and for which adaptive fitness might well be most important. Adaptive fitness is otherwise secured by heterozygosity in a wide range of organisms. The stabilizing influence of high genetic buffering against natural selection has been called genetic homeostasis by Lerner (1954). His definition of developmental homeostasis, or ontogenetic self-regulation, links it with the greater ability of heterozygotes to stay within the norms of canalized development. There is some evidence that developmental variability is complementary to genic variability in the sense that developmental variability increases where that of genetic origin is reduced (Scossiroli, quoted by Lerner, 1954). The canalization of Red and Desert Locusts takes the form of divergence between the developmental polymorphs in the early instars. On the basis of these divergent forms phase polymorphism is superimposed.

Subsequently, the twin growth-regulating mechanisms deflate the developmental polymorphism, of which the eye-stripes are relics, though the expressions of phase polymorphism persist. Indeed it is possible for a species such as *Anacridium aegypti* to show moulting polymorphism with few of the overt phase characteristics such as migration.

Red Locust egg-pods contain insects belonging to both developmental types. Isolated Locusts produce a preponderance of hatchlings subsequently to have 8 eyestripes, typical morphometrically when

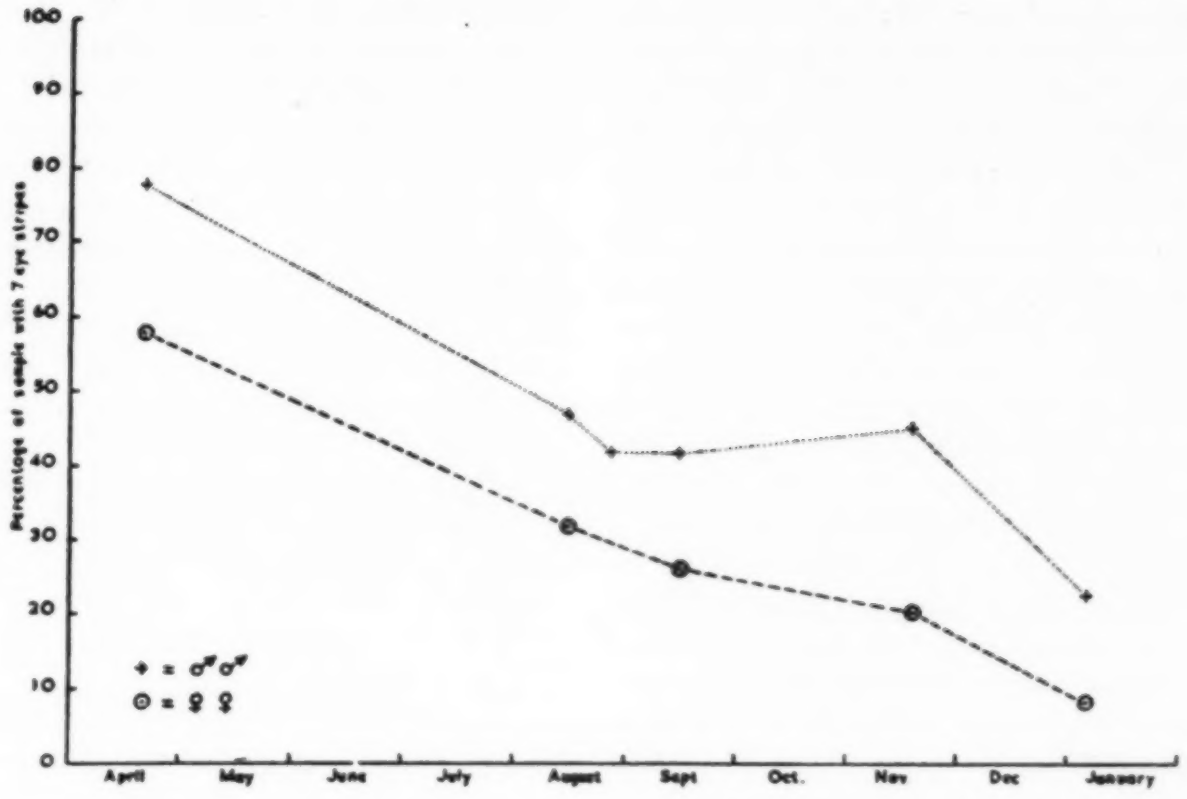

FIG. 5. Proportionate decrease of 7 eye-striped Red Locusts during 1952.

reared in isolation. One series of collections (fig. 5) throughout the dry season of 1952, from non-swarming populations of the single generation of Red Locusts in the Central Rukwa, indicated a progressive fall in the proportion of 6-instar forms in both sexes. Whilst the evidence of figure 5 is open to other interpretations we suggest that this is *prima facie* evidence of differential fitness and that relict forms are included in the product of each ovarial cycle even though one type may be in the minority.

Dobzhansky (1951) has noted that polymorphism within a species increases the efficiency of the exploitation of the resources of the environment, but the application of this idea in the present context is entirely speculative.

The extra moult occurs in Red Locusts, Desert Locusts, and occasionally in various sub-species of the Migratory Locusts (Nikolskii, 1925; Zolotarevsky, 1933; Remaudière, 1954). Volkonsky (1937) has found this extra moult, imprinted as an additional eye-stripe, in *Anacridium aegypti* but geographical differences obscure the extent to which this instance of polymorphism may be density dependent. In sparse populations of Red Locusts, two extra moults, with an imprint of 9 eye-stripes, are found in some individuals. Only one such individual has been bred in the laboratory, a reflection of the greater plasticity of field populations.

During 1948–9 when the Red Locust populations of the Rukwa valley were depleted, Burnett (1951) found a noteworthy proportion of 9 eye-striped insects. In 1950–51, the larval population density having increased, the proportion of such insects had diminished to about 1 per cent of those insects with additional instars (Albrecht, 1955).

Yet this developmental polymorphism is modified according to the sex of the locusts. Albrecht (1955) found that there were, in field samples of Red Locusts of a wide range of densities, consistently higher proportions of females which had undergone the extra moult. Nikolskii (1925) and Key (1926) found the extra moult only in a few female Migratory Locusts, and Zolotarevsky (1933) found it only in a few female *Locusta capito*. New light is thrown by Remaudière's (1954)

remarkable field investigation of the incidence of extra moults among female migratory locusts. In presenting this triple association between sex, moulting, and phase, we are trying to show how the delicate balance of polymorphism may be influenced throughout ontogeny by interwoven, probably hormonal, expressions of factors, some genically, some environmentally, inspired. This relation, between normal growth and phase status, is thrown back to a still earlier generation by recent work in collaboration with Michel Verdier: this work suggests that the weight at eclosion, and the maximum number of ovarioles which is fully determined at that time, of female Migratory Locusts are related to the phase status of their parents and, in turn, that these factors mediate the phase status of their offspring. Thus the influence of crowding persists through at least three generations.

Summary

The development of Red and Desert Locusts is considered in relation to phase and developmental polymorphism.

The recently discovered developmental polymorphs have different numbers of moults, characterised by the number of eye-stripes. Moreover, they differ in size, even when newly hatched, and for several instars the form eventually to have an extra instar, which is initially the smaller, grows more slowly than the other form, accentuating the initial difference.

The growth increment afforded by the extra instar removes most of this inequality of size, which is further reduced by the fact that within each polymorphic form, the initially larger insects grow less rapidly than those initially smaller.

There is little genic control of the proportion of the two forms among the progeny, but this proportion is closely linked with the integrated parental population density. As judged by the form of the wing pads and of the female genitalia, the form eventually to pass through the extra instar is morphologically typical only when reared in relative isolation, whereas the other form is typical of the *gregaria* phase. Attempts to rear either form at the density appropriate to the other give rise to individuals which can be classified as of *congregans* or *dissocians* phases. Analysis of phase reveals qualitative distinction from the normal processes of growth. The differences of specific form are distinct from size and phase differences; speciation is not here an exaggeration of variation latent in the polymorphic forms.

An attempt is made to discover the role of the developmental polymorphism in the ecology of locusts.

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EVOLUTIONARY TRENDS IN CRUSTACEA (MALACOSTRACA)

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The study of the phylogeny of the malacostracous Crustacea was based originally almost entirely on comparative morphology of living forms (Claus, Grobben, Calman), with important later contributions based on highly refined methods of functional analysis (Cannon, Dennell, Manton). Beurlen and Glaessner commenced some 25–30 years ago to confront the accumulated zoological evidence with palaeontological data. The evidence derived from fossils can be highly significant in this group, firstly because the Arthropod exoskeleton shows many important details of internal organisation, and secondly because the entire evolution of the Eumalacostraca proceeds in post-Cambrian time, before the eyes of the palaeontologist. In modern compendia and textbooks there is little evidence of the impact of the knowledge of fossil higher Crustacea on the reconstruction of their phylogeny. The classification used in treatises of palaeontology is mostly that established for living forms and many significant fossils are within the limitations of this system necessarily regarded as obscure and *"incertae sedis."* While some Palaeozoic Crustacea still deserve this designation until more is known about them, either from more detailed modern descriptions or additional discoveries, a review of others makes it possible now to give an outline of genetic relations between them and of their interpretation.

All Eumalacostraca are considered to be derived from an ancestor possessing the morphological characters described by Calman (1909, p. 144) as "caridoid facies." In Carboniferous time the representatives of different groups of Malacostraca, although to some extent distinguishable, were so close to each other and to a generalised ancestral type that little doubt remains about its real existence. Instead of the recent Mysidacea, Stomatopoda, Euphausiacea, Tanaidacea, we find Pygocephalomorpha, Perimecturidae, *Anthracocaris,* and other less well preserved forms, with a considerable number of common characters. It can be assumed that, but for our knowledge of their further differentiation into their recent descendants revealing the significance of their distinguishing characters, they would have been placed in a single Order of the Malacostraca.

The following genera should be included in the Order Pygocephalomorpha Beurlen (1930, p. 444) which was originally established for the genera *Pygocephalus* and *Crangopsis* only: 1. *Anthracophausia* Peach 1908, *Crangopsis* Salter 1863 (= *Palaeocrangon* Salter 1861 ***non*** Schauroth 1853), *Palaeopalaemon* Whitfield 1880. 2. *Pygocephalus* Huxley 1857 (Syn.: *Anthrapalaemon* Salter 1861, *Necroscylla* Woodward 1879), *Paulocaris* Clarke 1920, *Liocaris* Beurlen 1931, *Pygaspis* Beurlen 1934. 3. *Tealliocaris* Peach 1908, *Pseudogalathea* Peach 1882. These three groups differ mainly in adaptive characters and should be considered as families. Beurlen (1934, 1935) established for the genus *Pygaspis* a new Suborder Pygaspida, considering it as "benthonic descendants of the Marellidae or of a group closely resembling the Marellidae" (1934, p. 131, translated). His figures of *Pygaspis* resemble *Pygocephalus* very closely and I believe that the Brazilian Permian fossils described under this name represent the ventral structures of Malacostraca of the family Pygocephalidae. They resemble closely the similarly preserved *Notocaris tapscotti* Broom, most of which have the abdomen "folded round so that the telson lies under

the back part of the sternum" (Broom, 1931). This explains the peculiar outline of *Pygaspis*.

The primitive filter-feeding *Gnathophausia*-like Mysid from which the living benthonic Lophogastridae and pelagic Mysidae are believed to be derived (Manton, 1928, p. 116; Dennell, 1937, pp. 72–76) could well be represented by a Palaeozoic pygocephalomorph Crustacean very much like *Tealliocaris*, as Peach (1908) had already suspected. The Cumacea could have been derived from a similar form. Mysidacea and Euphausiacea were hardly distinguishable in Carboniferous time; the Recent Lophogastridae still resemble closely their common ancestors and the origin of the Decapoda which are not known from the Palaeozoic could not have been very different.

The primitive Schizopod characters of the Perimecturidae are actually the "caridoid facies" claimed by Calman to be an ancestral character of the Eumalacostraca. At the same time the Perimecturidae show clearly the evolutionary tendencies of the Stomatopoda, but there is no indication of convergence and there is nothing to exclude them from the ancestry of this Order. Grobben (1892) has in fact postulated a descent of the Stomatopoda from "primitive Schizopoda." Peach's view (1908, p. 53) was "that, while they are well within the Schizopod suborder, they may be looked upon as having taken on Squillid characters, and may even have been the early progenitors of the Squillid stock."

Peach put forward his views as expressed in the last words only hesitantly as an alternative because he based his conclusions on the characters of the recent Stomotopods without taking into consideration their probable evolutionary changes since the Palaeozoic. Incidentally, his views on the alleged Schizopod "habit of bending down the head and carapace on dying" and its taxonomic value are erroneous as they refer to the moulting position in which the cast shell becomes fossilised under favourable circumstances. It is frequently found in fossil Decapods (Glaessner, 1929, 1948).

The "ancestral Mysid" from which the Tanaidacea originated does not belong to the same group and could not have reached the level of the Pygocephalomorpha. A "greater gap" was observed by Dennell (1937) when he compared the relations between Apseudidae and Mysidacea with those between Cumacea and Mysids. This appears to be filled by the Acanthotelsonidae which could well include the postulated ancestral form with a small carapace and also provide the link between the most primitive caridoid filter-feeders and the later raptatory Isopods. The characters of *Anthracocaris* confirm the existence of the assumed phyletic line from the Acanthotelsonidae to the Tanaidacea. These relations are shown diagrammatically in the accompanying Table.

A review of evolutionary trends in Malacostraca shows that the surviving Divisions and Orders are by no means equivalent products of a simple process of adaptive radiation. Some of the existing groups can be described as "living fossils," such as the Nebaliidae, Syncarida and *Thermosbaena* which resembles *Perimecturus* remarkably closely. In the classification of the recent Malacostraca, this "living fossil" is the only representative of an Order Thermosbaenacea, believed to be related to the Stomatopoda. These living fossils owe their survival to combinations of exceptional ecological circumstances while many other forms which date as far back as these rare survivors have become extinct. In phylogenetic studies based entirely on the recent fauna such "living fossils" have often been considered as ancestral forms, with the unjustified exclusion of their well-known, more numerous, and often also more primitive and therefore more important *extinct* relatives.

Other groups such as the Mysidacea and Euphausiacea are conservative, showing a characteristic lack of functional differentiation. Cannon and Manton (1927, see also Manton, 1928) have shown that

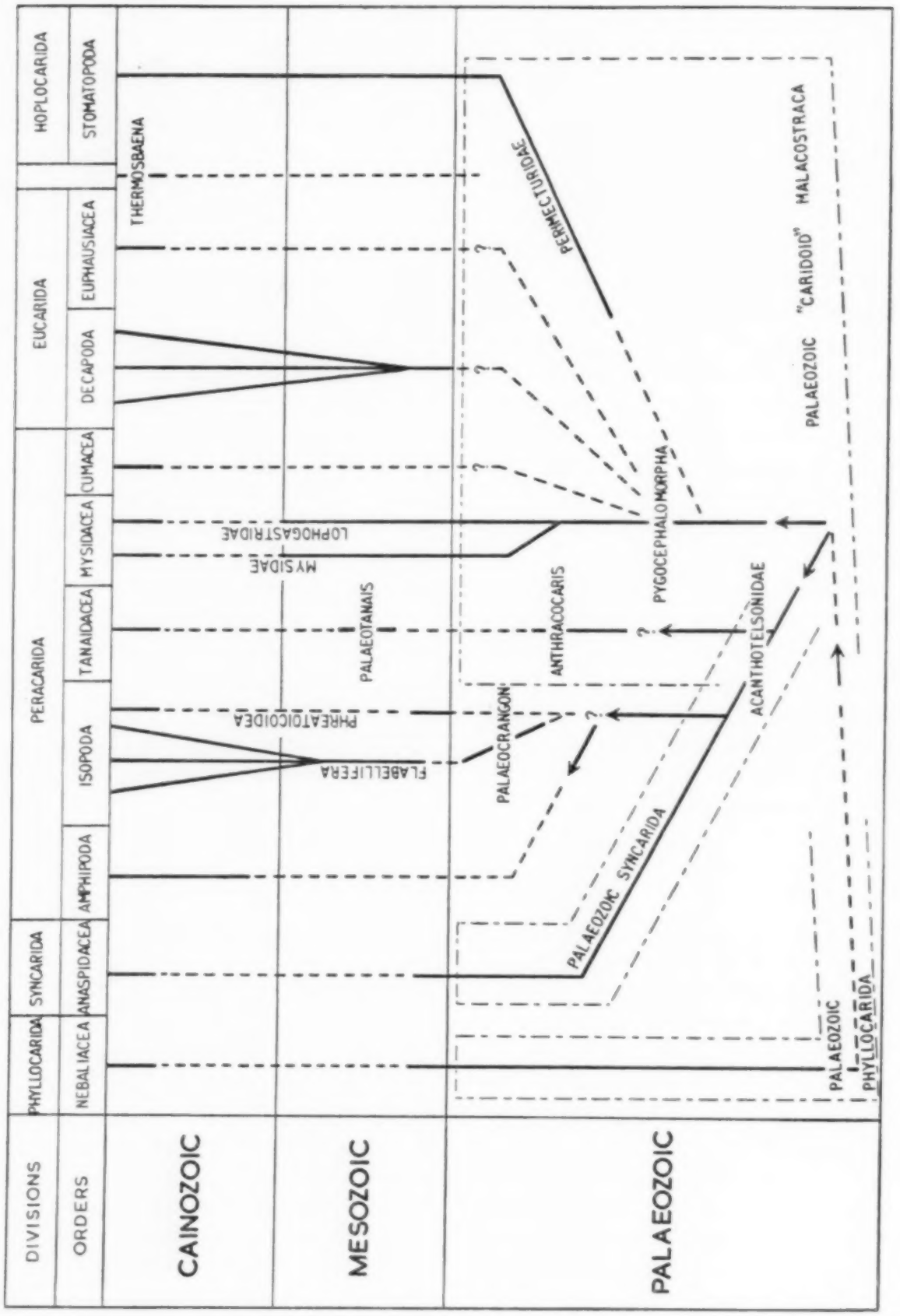

FIG. 1. Diagram of phylogeny of Malacostraca.

in the Mysidacea (s. str.) the functions of feeding, locomotion and breathing are inseparably connected with each other. Simultaneously and in a single cycle of movement the limbs produce, direct and utilise a stream of water for the purpose of breathing, feeding and locomotion. The structure of the limbs, their position, the arrangement of setae, etc., have become specialised, but instead of the development of distinct and separate organs there is a multiple use of a single organ. Similarly, the carapace participates in the functions of protection and breathing. Finally, the limbs contribute also to the reproductive equipment by developing oostegites. This reduced differentiation within the organism is clearly reflected in the lack of adaptive differentiation which distinguishes the Mysidacea from the Isopoda, Amphipoda or Decapoda.

In the Isopoda and Amphipoda, the carapace is gradually reduced and external segmentation of the body becomes almost perfectly uniform. With the exception of the parasitic forms which are morphologically aberrant and throw no direct light on the main lines of evolution these groups are well adapted to benthonic life. In the Isopoda, the endopodites of the thoracic limbs are the organs of locomotion, the exopodites having been lost. The branchiae are mostly formed as parts of modified pleopods. The original number of somites is frequently reduced, either by coalescence of abdominal segments or by fusion of anterior thoracic somites with the head. The general shape of the body is often dorso-ventrally depressed, as in trilobites.

In the Amphipoda there is likewise no carapace and the body is more or less uniformly segmented. The limbs, however, are more strongly differentiated. Branchiae are formed on the thoracic limbs. The exopodites are absent. The coxopodites of the thoracopods form branchial plates and oostegites. Articulations and movements are such that the breathing is separated from locomotion, and there is no direct connection between the movement of the thoracic legs and the function of the gills. The body is mostly laterally compressed. The normal method of locomotion is crawling or leaping, either in normal position or lying on one side. Dorso-ventrally compressed forms with reduced abdomen climb on plants. The ecologic and geographic range of the group is very considerable and the morphological and taxonomic differentiation of the Amphipoda is intense.

The evolution of the two lineages of Peracarida which have lost the carapace and acquired a complete segmentation and benthonic mode of life can be considered as progressive. In both groups the functions of protection, locomotion, feeding and breathing are separated. The differentiation of the two parallel lineages (Isopoda and Amphipoda) is probably geologically old (Glaessner, 1957) but palaeontological data so far fail to throw any light on its origin. Morphological evidence of their early differentiation from the ancestral caridoid stock was discussed by Calman (1909, p. 240).

The most significant morphological character of the Stomatopoda is the progressive development of the abdominal portions of the body concurrently with the general reduction of the carapace. Organs migrate into the abdomen which becomes extremely large and strongly developed. Even the branchiae are formed on the pleopods instead of the thoracic limbs. This relieves the thoracopods of their participation in the breathing function. The shifting of the gills and of the mechanical centre of gravity makes the exopodites superfluous; the endopodites take over the function of locomotion. The mobility of the anterior somites of the cephalothorax can be increased, as important internal organs are moved into the abdomen. This, together with the retreat of the carapace from the posterior thoracic somites and its splitting up into separate plates makes it possible for the anterior thoracic limbs to become raptorial. The diminished protection resulting from the reduction of the carapace is

compensated to some degree, as in some anomurous Decapods, by burrowing mode of life, and also by extremely rapid movement effected by the strong muscles in the abdomen. The evolutionary change from a primitive "caridoid" Malacostracan through the fortunately preserved intermediate stage of *Perimecturus* to the morphologically and ecologically aberrant true Stomatopoda (Hoplocarida) is essentially a process of far-reaching internal and ecological adjustment to a comparatively minor primary divergence.

A truly progressive evolution occurs in the Decapoda, leading far beyond the original level of organisation and greatly increasing the range of possible adaptation. Evolution in the Decapoda is clearly directed towards differentiation and increase in functional importance of the carapace and the thoracopods.

The carapace, originally in the Phyllocarida little more than a protective fold for internal organs, enables the organism to achieve a vast improvement in its branchial apparatus and in the respiratory mechanism as soon as it replaces the original thoracic integument which becomes redundant under the new cover. The carapace is completely fused with the thoracic somites, forming a branchial chamber in which a constant current is maintained. Only the gnathocephalic appendages participate in its production. Oostegites are absent, the development is free but the eggs are carried on the pleopods. In the Decapoda those thoracopods which are not modified to maxillipeds are free from all functions other than locomotion and active taking of food. Further evolution which produces a remarkably wide ecologic range and variety of form is closely related to elaboration of the morphological basis of the locomotive function.

In this connection the reduction of the abdomen is particularly significant. As long as its appendages, the pleopods, act in conjunction with thoracopods as swimming paddles it is a normally functioning organ, as in lower Malacostraca. Where thoracopods take over the propulsion, its only participation in locomotion is by sudden flexion producing a strong backward flight movement. As the abdomen is incapable of developing walking legs comparable with those of the thorax, it must impede the normal mobility of benthonic animals possessing a rigid cephalothorax.

Three different trends are observed in the evolution of the Decapods. One produces swimming forms (Natantia), apparently by neotenic (paedomorphic) processes, on two or more different levels (Penaeidea, Caridea). Another trend is conservative, retaining the division into cephalothorax and abdomen in benthonic forms (Nephropsidea). It is a striking fact that these forms which were very common and highly differentiated in Jurassic time become much less numerous in individuals and taxonomic divisions in late Mesozoic and Cainozoic time (van Straelen, 1936). At the same time, in a number of other groups and along various lines a tendency appears towards reduction of the abdomen and elimination of its locomotive function. A *reduction in calcification* of the integument drives the Thalassinidea and Paguridea into limited ecological niches where external protection (by burrowing in the ground or concealment in molluscan shells) assured survival from the Jurassic to the present time at the cost of enforced structural alterations seriously reducing the possibilities of conquering wider fields. A *reduction in size* of the abdomen *involving a loss* of important thoracic skeletal structures produced in the Galatheidea a small number of "pseudo-Brachyura" (Porcellanidae). Only one method, a reduction in relative size of the abdomen coupled with gradual but efficient *reconstruction of the internal skeleton* of the thorax become spectacularly successful. As soon as initial difficulties in internal adjustments were overcome and the stage of the Dromiacea which was dominant throughout the Jurassic was left behind, the Brachyura became extremely nu-

merous, widespread, and highly differentiated. The functional (but not morphological) removal of the impeding abdomen in the Brachyura made them not only better adapted to the habitat of their Macruran ancestors but as soon as all enforced internal readjustments were completed, it opened the path to vast improvements in the mechanism of locomotion. The ecologic range of the new type was then extended to the littoral and even terrestrial habitat.

Conclusions

The early Malacostraca had inherited a differentiation of the integument into carapace and abdomen. The first major steps in their further evolution follow two divergent paths: one toward consolidation of that differentiation, the other back to uniform segmentation as far as this was possible on the malacostracan level of evolution. The primitive representatives of this branch are the Syncarida (which have survived to the present time); their more advanced descendants are the Isopoda and Amphipoda. The other major groups of Malacostraca either show remarkably little functional differentiation (Mysidacea) and correspondingly no significant adaptive radiation, or they indicate aberrant tendencies (Cumacea, Stomatopoda), or they pass through the stage of consolidation of the division between carapace and abdomen to a further stage in which the abdomen is reduced so that it and its appendages take no part in locomotion and have few other functions. In other words, external segmentation almost disappears. The evolution of the Decapod Crustacea from their first appearance at the beginning of the Mesozoic Era to their extreme present diversification and abundance can be followed with the aid of abundant fossil material but details are beyond the scope of this contribution. The three different ways of the reduction of the abdomen (one of which only is "successful" in the Brachyura), taken together with the decline of the Macrura Reptantia since the Mesozoic indicate strong selection pressure against the retention of the two-fold division of the body in benthonic Malacostraca. This observation has a bearing on the interpretation of the aberrant Stomatopoda in which the primary division is largely overcome in the opposite way, in favor of the abdomen. Their known Palaeozoic ancestors show incipient specializations in this direction though they are still close to the ancestors of the Decapoda and Mysidacea in other characters.

The fact that the classification of living organisms is an inadequate framework for the study of their genetic relations and that conclusions drawn from their comparative and functional morphology and embryology, though potentially valid, need corroboration from fossils and their distribution in time is well known from other groups but has not yet been made sufficiently clear for the Malacostracous Crustacea. When freed from the shackles of the horizontal classification of the living forms, the fossil material can be evaluated. Lines of descent followed (in a first approximation) from Palaeozoic to Recent times indicate two facts of possibly general significance.

(1) The living malacostracous Crustacea are heterogeneous results of (a) "successful" evolutionary trends, with strong adaptive radiation and dispersal to previously inaccessible zones or habitats, probably including examples of tachytelic evolution, (b) undifferentiated lines, probably bradytelic, (c) survival of "living fossils" in special ecologic niches or in geographic isolation. There is no evidence compelling us to consider them, as Beurlen has done, as the result of one or two pronounced "explosive" evolutionary phases in the Carboniferous and Permian.

(2) The dominant "theme" or "problem" in the evolution of this group is the inherited division of the body into an externally unsegmented anterior and a more or less isometameric posterior portion, with corresponding differentiation (functional or structural) of appendages into

two groups which must be coordinated. Locomotion as well as other functions can be perfected only when this is overcome. The two "successful" phyletic groups obviate this division in the two basically possible ways, by an approach either to complete secondary isometamerism or to complete "concentration" with loss of external segmentation. Evolution is here as always opportunistic rather than orthogenetic and various possibilities are being realised in the course of time on different levels of organisation. Original divergences are generally minor (as seen within the Pygocephalomorpha, between them and the Perimecturidae, or in the early Brachyura) and only when the prolonged period of internal adjustment made necessary by the first step in the reduction of either carapace or abdomen is completed they are seen to be far-reaching and to be followed by rapid adaptations to new habitats. It is doubtful whether this case of adaptation following modification of an impeding or retarding inherited basic structure is akin to preadaptation; selection certainly brings about both series of steps but the difference between the early stage and the subsequent adaptations made possible by it should be noted.

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VARIATION IN FERTILITY OF HYBRIDS BETWEEN ISOLATED POPULATIONS OF THE SERPENTINE SPECIES, *STREPTANTHUS GLANDULOSUS* HOOK

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The outcrops of serpentine and other ferromagnesian rocks in California are well known for the array of distinctive endemic species that their derived soils support (Mason, 1946; Stebbins, 1942). The causal aspects of restriction to serpentine, the chemical basis of the edaphic endemism, the tolerance of certain plant species to the low nutrient status of Californian serpentine soils, and other aspects of this striking form of endemism are discussed in Kruckeberg, 1951, 1954; Walker, 1954, 1955; and Whittaker, 1954.

The restriction of a number of species of the cruciferous genus Streptanthus to serpentine soils of California has been of particular interest to students of plant geography and evolution (Kruckeberg, 1951, 1954; Mason, 1946; Morrison, 1941; Stebbins, 1942; Walker, 1954). Some species of Streptanthus are found on but one or two serpentine outcrops of only a few miles in extent. Others may show wider distribution but remain obligate to the serpentine habitat. Still others may be restricted chiefly to serpentine but in addition possess populations intolerant of serpentine (Kruckeberg, 1951). Such facultative endemism to serpentine is exhibited by *Streptanthus glandulosus* Hook.

Streptanthus glandulosus, a species complex of many phenotypically distinguishable strains, is widely distributed throughout the Coast Ranges of central and northern California. However most populations are spatially isolated from one another due to the discontinuity of suitable habitats (mainly serpentine outcrops). Several of these local populations throughout the range of the complex have been considered distinct enough to merit taxonomic recognition as species. Such a network of edaphically isolated and morphologically distinct populations poses problems in evolution and taxonomy that are amenable to experimental attack. This paper presents results of hybridizations involving thirty-two different population samples. Plants grown from seed collected in the wild served as the pollen and seed parents of over 300 artificial hybrid combinations. The major effort has been to determine and evaluate any correlation between the extent of spatial isolation of the various populations and the degree of fertility of their hybrids. In addition, study of meiosis in many of the hybrids has been pursued in an attempt to establish a cytological basis for the variation found in the fertility of interpopulational hybrids.

This analysis of hybrid fertility thus has led to an assessment of the magnitudes of genetic isolation that may exist between spatially isolated populations. On the premise that degree of genetic isolation is correlated with degree of taxonomic relationship, the results have been applied to a taxonomic interpretation of the complex. Intrinsically associated with the taxonomic objective is an evolutionary one: To achieve an understanding of the *kind* of genetic isolation that has developed during the elaboration of the polymorphism exhibited by the group.

B and C of Fig. 2 facing

FIG. 2. Inflorescences of the five taxa in the *Streptanthus glandulosus* complex.

A row: *S. niger*, *S. albidus*-MD and MR. B row: *S. glandulosus* subsp. *glandulosus*-CD, and MRC, subsp. *pulchellus*-BJ, and *S. albidus*-BH. C row: *S. glandulosus* subsp. *secundus*-CZ, MJ, and KV, and *S. glandulosus* subsp. *glandulosus*-RG. Letters following each taxon are for strains listed in table 2. Photographs by the Still Photo Unit, University of Washington.

B

C

FIG. 5. Inflorescences of the hybrid (center) between *Streptanthus glandulosus* subsp. *glandulosus*-MRC (left) and *S. glandulosus* subsp. *secundus*-OC (right), (see p. 191).

The two abbreviations designate strains listed in table 2. Photographs by the Still Photo Unit, University of Washington.

siderably the limits of Hooker's *S. glandulosus*. However in the past, less conservative taxonomists accorded species status to a number of these color morphs. In addition to *S. glandulosus*, no less than twelve species were recognized by E. L. Greene, eleven of which he placed in another genus Euclisia (Greene, 1906). Jepson (1925) and Abrams (1944) saw only three species in the complex: *S. glandulosus* with two varieties (var. *albidus* and var. *pulchellus*), *S. niger* Greene and *S. secundus* Greene. Morrison (1941), in the only recent monographic treatment of *S. glandulosus*, places this species and one other in the subsection Pulchelli of the section Euclisia. To four of the species recognized by Greene (*S. albidus, S. niger, S. pulchellus* and *S. secundus*), Morrison has given subspecific status under *S. glandulosus*. The most widespread element in *S. glandulosus* was designated by Morrison as subspecies *typicus* (= subsp. *glandulosus* of the present author). *Streptanthus Coombsae* Eastwood, the other species recognized by Morrison, apparently has not been collected since its discovery in southern Oregon in 1913 and hence will not be dealt with here.

None of the earlier taxonomic treatments mirrors the polymorphism of the

TABLE 1. *Synopsis of taxa in the* Streptanthus glandulosus *complex*

Taxon	Distinguishing features	Distribution
S. glandulosus subsp. *glandulosus*	Flower color ranging from rose, lavender, violet, purple, to purplish black. Infl. a non-secund raceme. Plants dwarfish to robust; sparingly pubescent to hispid.	Coast Ranges from Mendocino to northern Santa Barbara Counties, "North Coast" group from Mendocino and Sonoma Counties with rose-colored flrs; "North Interior" group from inner Coast Range areas with dark purple to blackish flrs; "South Coast" group from Santa Clara to southern Monterey Counties with dark purple flrs; those from San Luis Obispo County with lilac flrs. Discontinuous mainly on serpentine in all counties within this region.
subsp. *pulchellus*	Flower color reddish purple; infl. usually secund; plts. often dwarfish.	Marin County serpentines, especially on slopes of Mount Tamalpais. Distinguished from Marin County subsp. *secundus* only in flr. color.
subsp. *secundus*	Flower color yellow to creamy white; petals occasionally veined with violet; infl. secund.	Marin and Sonoma Counties, predominantly on serpentine; Jackson County, Oregon plants probably referable here. "Marin County" plants with greenish-white calyces and yellowish petals, veined with violet; "Sonoma County" plts. with flrs. either pure yellow or greenish-white.
S. albidus	Flower color lilac to greenish white. Plants taller, more robust; glabrous throughout.	Alameda and Santa Clara Counties, mainly on serpentine. Flrs. of BH, SC, and MD strains are pale lilac; those of MR plants are greenish white.
S. niger	Flower color purplish black. Corolla constricted at throat causing anthers and style to be congested at anthesis. Infl. "zig-zag"; pedicels twice as long as flrs. Plants wholly glabrous.	On serpentine, Tiburon Peninsula, Marin County.

TABLE 2. *Origin of strains used in hybridizations*

Taxon and region	Designation of strain	Locality and rock type
S. glandulosus subsp. *glandulosus*		
North Coast	NV	Navarro River, Sonoma County. *F. W. Hoffman.*
	RG*	Russian Gulch, Sonoma County, Basalt.
North Interior	CD*	Conn Dam, Napa County. Serpentine.
	CM	Contact Mine area near Mt. St. Helena, Sonoma County. Serpentine.
	GY	The Geysers, Sonoma County. Serpentine.
	KX*	One mile west of Knoxville on road to Lower Lake, Lake County. Serpentine.
	LK*	Highland Springs, Lake County. Serpentine. *F. W. Hoffman.*
	MO*	Between Longvale and Covelo, Mendocino County. *F. W. Hoffman.*
	MRC*	Near Mercuryville on road to The Geysers, Sonoma County. Serpentine.
	PO*	Pope Creek Canyon near Walter Springs, Napa County. Serpentine.
	WC*	Upper Weldon (Mix) Canyon, Vaca Mountains, Solano County. Sedimentary.
	1030*	Three miles northeast of Middletown, Lake County. Serpentine.
South Coast	MY*	Nacimiento Creek, Monterey County. *F. W. Hoffman.*
	SLO*	Between Paso Robles and Cambria, San Luis Obispo County. Serpentine. *F. W. Hoffman.*
S. glandulosus subsp. *secundus*		
Marin County	LV*	At head of Lucas Valley. Serpentine.
	SG*	On San Geronimo Ridge. Serpentine.
Sonoma County	CZ*	Between Cazadero and Richardson's Grove. Shale.
	DT	Dutch Bill Creek, near Occidental. Serpentine. *F. W. Hoffman.*
	GB*	Gabe's Rock, along Poole Ridge, just nw. of Guerneville. Serpentine.
	KV*	Upper Knight's Valley. Shale.
	MJ*	Mt. Jackson mine near Guerneville. Serpentine.
	MK	Just east of Camp Meeker. Serpentine.
	OC	Just east of Occidental. Serpentine.
	SW*	Sweetwater Springs road, just east of Mt. Jackson mine. Serpentine.
Josephine County, Oregon	ORE*	Northeast of Takilma. Serpentine.
S. albidus Greene	BH*	East slopes of Berkeley Hills, near Orinda tunnel, Alameda County. Basalt.
	MD*	Two miles inside old north entrance to Mt. Diablo State Park, Contra Costa County. Serpentine.
	MR*	Metcalf Road, one mile east of Coyote, Santa Clara County. Serpentine.
	SC*	Upper Stevens Creek, Santa Clara County. Gabbroic rock.
S. glandulosus subsp. *pulchellus*	BJ*	Bootjack Camp, Mount Tamalpais State Park, Marin County. Serpentine.
	RC*	Rifle Camp, Mount Tamalpais State Park, Marin County. Serpentine.
S. niger Greene	TB*	Southern end of Tiburon Peninsula, Marin County. Serpentine.

* Denotes the gametic chromosome number of n = 14 determined for this strain.

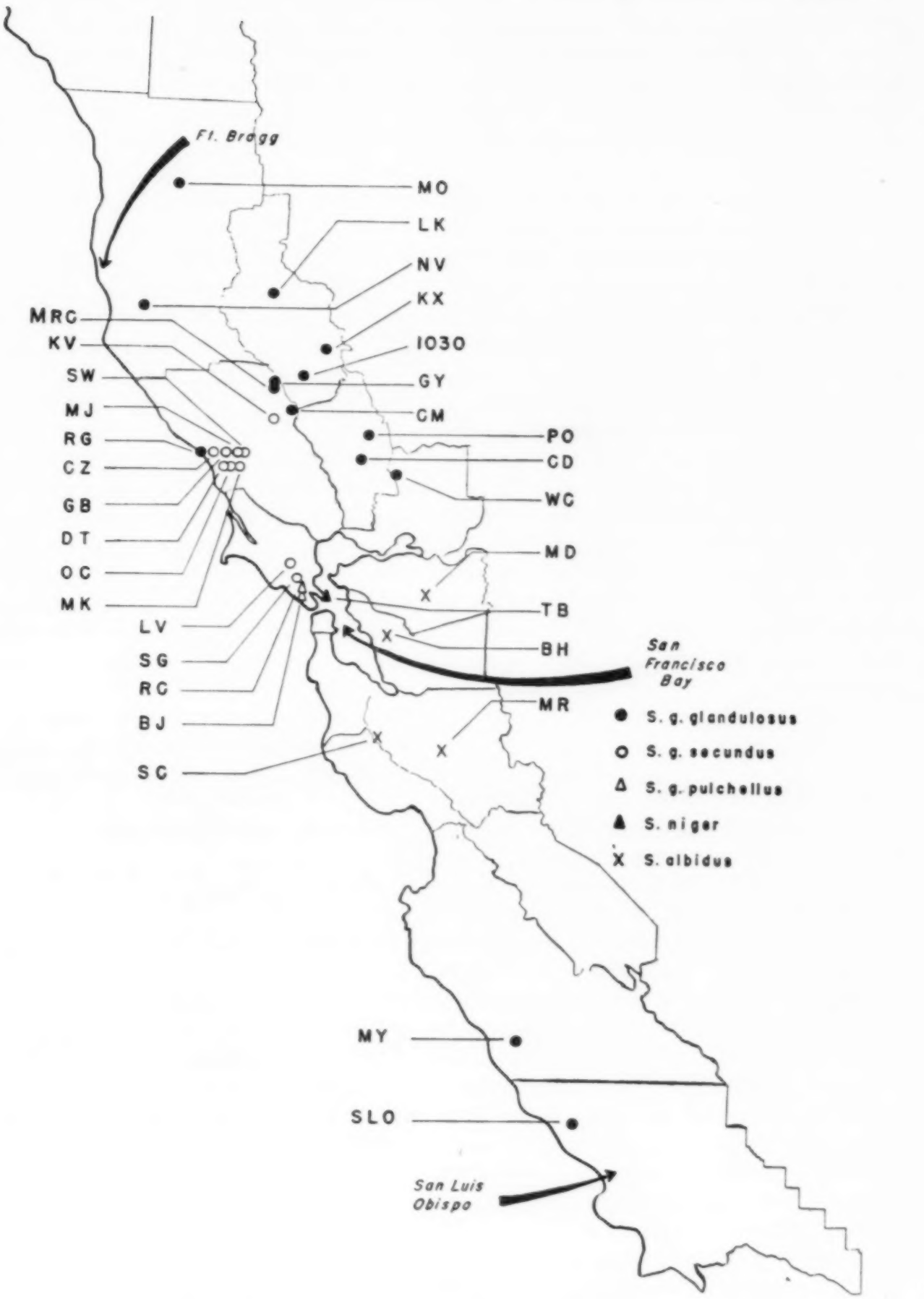

FIG. 3. Map of the Coast Ranges of California showing the distribution of the 32 strains of the *Streptanthus glandulosus* complex used in the hybridizations.

See table 2 for details. Abbreviations designate strains listed in table 2.

group that is seen in nature, although Morrison's study is the most satisfactory to date. Taxonomic assessment of the complex, based on the results of the present study of interstrain fertility, may better reveal the biosystematic status of the group.

Since reference will be made throughout this paper to the taxa comprising the complex, it seems advisable to present a taxonomic summary at this point. A synopsis of the five taxa in table 1 represents the author's disposition of the major segments of the variation pattern. These five taxa are given status as species or subspecies according to the degree of genetic isolation that has been achieved by the constituent populations. The criterion for "degree of genetic isolation" has been the level of pollen fertility of artificial interpopulational hybrids, as documented in the "Results" section of this paper. Formal publication of the taxonomy of the group is planned as a separate paper.

Materials and Methods

Plants of *Streptanthus glandulosus* Hook., employed in the hybridization program, were grown from seed collected in the field. Table 2 gives the source, chromosome number, and taxonomic disposition of the collections serving as parents for the crosses. Hereafter the various strains will be referred to by the abbreviations listed in the table. The distribution of the 32 strains is shown in figure 3; it is plotted according to the taxa recognized by the author.

In making the crosses, flowers were emasculated and 24–48 hours later were pollinated; the entire operation was performed in insect-proof cages. The hybrid seed was sown the following season. The F_1 plants provided the results to be presented in this paper. Bud samples for cytological analysis, pollen samples for fertility counts, and herbarium vouchers were taken from parental and hybrid plants. Photographs of single flowers and of entire plants were taken of representative parental strains and hybrids. Buds were fixed in a solution of three parts 95 per cent ethyl alcohol to one part glacial acetic acid. Meiosis in pollen mother cells was studied by means of squashes of anthers in iron-acetocarmine. Staining was enhanced by dipping the squashing implements in a 10% acid solution of ferric acetate.

Fertility of parental strains and hybrids was estimated from counts of 200 pollen grains stained in aniline blue-lactophenol. The sample size of 200 was selected on the basis of preliminary counts of 100, 200, 300, and 500 pollen grains; it was found that for a given sample the per cent of stainable pollen did not vary significantly after 200 grains were counted. Each count was made on freshly collected pollen from a single flower just prior to exsertion of the anthers. A minimum of two hybrid plants was sampled; however in most cases, pollen from four to six different plants was counted and an average recorded as the final count. Plump, fully stained pollen grains were considered to be viable.

Results

Pollen Fertility and Meiotic Behavior of Parental Strains

Counts of fresh pollen were made for each of the strains used as parents in the interpopulational hybrids. All parental strains showed good pollen fertility (95–100 per cent stainable pollen). High pollen fertility, coupled with the observation of good seed yields from artificial self-pollinations indicated that the parental plants are fertile.

Fourteen bivalents are consistently found at diakinesis of meiosis in pollen mother cells (fig. 4a and d). No evidence of irregularity was seen in the subsequent stages leading to pollen formation (fig. 4b and c). Those strains for which chromosome numbers have been determined are designated by an asterisk in table 2.

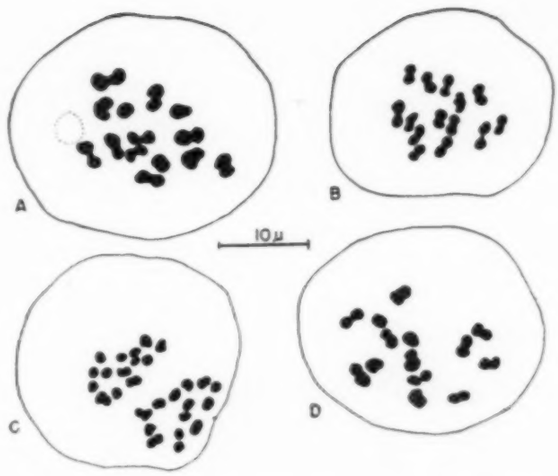

FIG. 4. Meiotic chromosomes of *Streptanthus glandulosus*. Figs. 4a-c. Diakinesis, M_I, and M_{II} for the SW strain of *S. glandulosus* subsp. *secundus*. Fig. 4d. Diakinesis for the MRC strain of *S. glandulosus* subsp. *glandulosus*.

POLLEN FERTILITY OF THE F_1 HYBRIDS

Artificial hybridizations between any two strains of the *Streptanthus glandulosus* complex almost invariably yield viable F_1 seed. The vigorous F_1 progeny are distinguished from their parents in displaying intermediate aspects of flower color, morphology, and habit. Figure 5 shows a typical hybrid, flanked by its parents, *S. glandulosus* subsp. *glandulosus* (MRC) and *S. glandulosus* subsp. *secundus* (OC). The markedly reduced pollen fertility of many of the hybrids made it apparent that the hybrids could not be regarded simply as recombinants within a panmictic population complex. Of the 334 (out of a possible 992—i.e., $32^2 - 32 = 992$) F_1 combinations that were grown, almost half were significantly less fertile than their parents. The criterion of fertility (stainability of pollen) often only served to confirm the infertility of the hybrid which was at once obvious from an inspection of the shrivelled, aborted anthers. Those hybrids in which anthers were malformed were the least fertile; plants of this type seldom produced more than 10 per cent stainable pollen and mostly less than 5 per cent stainable pollen.

The fertility of the F_1 crosses is presented in table 3 as per cent of stainable pollen. The data of the table are arranged as follows: Each parental strain is designated by its abbreviation as listed in table 2. The first of a pair of parental strains served as the seed parent. The hybrid combinations are grouped according to the taxonomic and geographic designation of the seed parent. Within each such group the crossing combinations are arranged in order of increasing distance in miles that separates seed and pollen parent. Each per cent value represents an average of two or more counts of 200 pollen grains from each of two flowers from different hybrid plants.

Since the evidence for relative fertility of interstrain hybrids comes primarily from counts of stained pollen, the reliability of the method must be considered. Two sorts of variability that may impair the accuracy of this type of data are encountered. First, different plants of the same F_1 hybrid population may differ in pollen stainability, rarely by as much as 20 per cent. Secondly, the same plant when sampled at different times may give somewhat different values for pollen stainability. When such variations were encountered, additional samples were counted until satisfactory agreement between successive counts was obtained. Thirdly, differences between the pollen stainability of a hybrid and that of its reciprocal were noted. It is because of these limitations that the per cent values have been rounded off to the nearest 5 per cent.

The uniformity of sampling and counting partially compensates for the above limitations. The author took all pollen samples, ensuring a uniform procedure for sampling. Mr. Robert Ornduff made the counts for 1956, using a compound binocular microscope at × 40 magnification. One ocular of the inclined binocular body tube was completely blocked off and in the ocular of the other barrel, a black celluloid disc with a 2 mm square opening in the center was inserted; this contrivance reduced the field to a smaller,

TABLE 3. *Pollen fertilities and intervening distances for each interstrain hybrid* *

Interstrain hybrid	Per cent good pollen	Distance between strains
S. albidus as seed parents		
BH × other *S. albidus*		
BH × MD	95	25 miles
× SC	60	40
× MR	90	50
BH × other strains		
BH × TB	0	15
× LV	0	30
× OC	0	60
× GB	0	60
× KV	0	65
× CM	0	65
× GY	0	70
× MRC	0	70
× 1030	10	70
× CZ	0	70
× RG	0	75
× LK	0	90
× MY	0	135
MD × other *S. albidus*		
MD × BH	90	25
× MR	100	50
× SC	95	50
MD × other strains		
MD × TB	10	30
× BJ	0	45
× RC	0	45
× SG	0	45
MD × LV	0	55
× KV	0	75
× CM	0	75
× 1030	0	75
× MJ	0	80
× GY	0	80
× MRC	0	80
× GB	0	85
× NV	0	125
× SLO	25	160
× ORE	0	290
MR × other *S. albidus*		
MR × SC	65	20
× BH	100	50
× MD	95	50
MR × other strains		
MR × TB	50?	60
× RC	0	70
× RG	0	125
SC × other *S. albidus* & others		
SC × MR	95	20
× BH	55	40
× MD	95	50
× CD	5	85
S. g. subsp. glandulosus (*North Coast*) *as seed parents*		
RG × CZ	100	5
× SW	100	15
× GY	90	30
× MRC	90	30
× CM	95	30
× LK	95	50
× LV	80	55
× RC	50	60
× SC	0	110
NV × RG	95	40
× GY	95	45
× MJ	95	45
× MO	0?	45
× KV	80	55
× CM	80	55
× 1030	85	55
× RC	35	100
× SLO	0	270
S. g. subsp. glandulosus (*North Interior*) *as seed parents*		
CD × 1030	0	30
× KX	75	35
× MK	30	40
× CZ	5	45
× SC	10	85
CM × MRC	90	10
× KV	95	10
× GY	95	10
× KX	75	20
× RG	70	30
× MY	0	200
GY × KV	90	10
× CZ	95	25
KX × 1030	50	15
× CD	90	35
× WC	90	45
× RG	0	50
LK × 1030	100	20
× MRC	100	30

* See table 2 for meanings of strain abbreviations in the left-hand column. The seed parent of a cross is given to the left of the crossing symbol (×); the male parent to the right. Within each taxonomic and geographic group of seed parents, the hybrid combinations are arranged in order of increasing distance in miles intervening between strains serving as seed and pollen parents.

TABLE 3.—*Continued.*

Interstrain hybrid	Per cent good pollen	Distance between strains
S. g. subsp. glandulosus.—Continued.		
LK × CM	95	30
× KV	100	35
× RG	95	50
× CZ	95	50
× GB	90	50
× SW	90	50
× OC	95	55
× MO	100	55
× BJ	65	85
× RC	40	90
× TB	10	90
× BH	5	90
× MD	0	100
× SC	0	140
× MY	0	230
× SLO	0	250
MO × NV	95	45
× LK	100	55
× MRC	90	70
× GY	75	70
× KX	45	70
× KV	85	80
× CM	95	80
× RG	100	85
× MK	15	85
× WC	5	120
× TB	10	140
× MD	0	150
× SC	0	190
× MY	10	300
× SLO	0	330
MRC × KV	95	10
× 1030	85	15
× SW	85	20
× LK	95	30
× OC	50	30
× SG	50	60
× MO	90	70
× MD	0	80
× ORE	50	300
PO × WC	0	20
× 1030	95	25
× KX	0	25
× KV	95	25
× GY	100	30
× MRC	95	30
× SW	95	40
× LK	95	45
× SG	75	45
× RG	45	50
× MO	95?	95
× SC	0	95
× MY	0	190
× ORE	65?	250
S. g. subsp. glandulosus (South Coast) as seed parents		
MY × MD	0	135
× BH	0	135
× TB	10	140
× SG	0	150
× KV	85?	200
× MO	15	300
× ORE	75?	435
SLO × MY	5	30
× MD	25	160
× TB	0	170
× CD	0	200
× GB	0	225
× KV	0	225
× 1030	0	230
× MRC	0	230
× RG	0	230
× KX	0	235
S. g. subsp. secundus (Marin County) as seed parents		
LV × SG	85	5
× OC	95	35
× MK	75	35
× CZ	95	50
× GB	95	50
× KV	80	50
× MJ	90	50
× RG	75	55
× MD	0	55
× 1030	5	55
× MR	0	75
SG × RC	95	<5
× LV	95	5
× BJ	95	10
× OC	85	40
× MD	0	45
× MJ	55	50
× GB	35	50
× DT	65	50
× SW	35	50
× KV	60	55
× CZ	60	55
× CM	45	55
× RG	70	55
× SC	0	60
× LK	60	80
× MO	65	130
× ORE	50	300
S. g. subsp. secundus (Sonoma County) as seed parents		
CZ × GB	100	5
× MJ	100	5
× SW	95	5

TABLE 3.—*Continued.*

Interstrain hybrid	Per cent good pollen	Distance between strains
S. g. subsp. secundus.—Continued.		
CZ × RG	100	5
× OC	95	15
× KV	90	20
× LV	70	50
× SG	85	55
× RC	70	55
× BH	0	70
DT × MJ	95	5
GB × CZ	100	5
× MJ	100	5
× RG	100	10
× MRC	90	25
× 1030	95	30
× BJ	85	50
× SG	55	50
× ORE	90?	250
KV × CM	95	10
× GY	95	10
× 1030	90	15
× MJ	100	20
× CZ	95	20
× DT	85	20
× GB	85	20
× SW	95	20
× OC	70	25
× RG	95	30
× WC	0	45
× LV	30	50
× SG	50	55
× BJ	80	55
× RC	70	55
× TB	20	55
× BH	0	65
× MD	0	75
× SLO	0	225
MJ × SW	95	<5
× GB	95	5
× DT	95	5
× KV	95	20
× MRC	95	20
× 1030	85	35
× PO	15	45
× LV	70	50
× SG	70	50
× RC	70	55
× BJ	40	55
× MO	95?	85
× MY	0	200
OC × MK	90	<5
× MJ	95	10
× GB	90	15
× CZ	100	15
× RG	100	15
× KV	70	25
OC × 1030	80	30
× LV	95?	35
× KX	0	45
× TB	0	50
× BH	0	60
× SLO	0	225
SW × MJ	95	<5
× GB	100	5
× DT	95	5
× OC	95	15
× KV	95	20
× CM	95	20
× GY	90	20
× 1030	95	30
× PO	95	40
× CD	0 (40)	40
× LV	80	45
× LK	95	50
× SG	65	50
× RC	85	50
× NV	65	55
× BJ	65	55
× MO	95?	85
× SC	0	105
× MY	0	200
× ORE	45	250
S. g. subsp. secundus (Josephine County, Oregon) as seed parents		
ORE × LK	70	210
× 1030	55	230
× RG	80	250
× CM	70	250
× CZ	80	250
× GB	70	250
× OC	90	260
× MD	0	290
× TB	0 (65)	290
× RC	100?	300
× SG	0	300
× SC	0	340
S. niger as seed parents		
TB × SG	0	10
× WC	0	40
× OC	0	50
× KV	5	55
× MR	10	60
× 1030	0	65
× KX	0	75
× LK	10	90
S. g. subsp. pulchellus as seed parents		
BJ × RC	100	<5
× TB	0	5
× SG	95	10

TABLE 3.—*Continued.*

Interstrain hybrid	Per cent good pollen	Distance between strains
S. g. subsp. pulchellus.—Continued.		
BJ × LV	100	15
× OC	90	45
× MD	0	45
× MJ	80	50
× GB	95	50
× SW	30	55
× GY	75	55
× RG	60	55
× SC	0	55
× CM	40	55
× KV	55	55
× 1030	80	60
× MR	0	75
× LK	60	85
× NV	70	95
× MY	0	150
× ORE	100	300
RC × SG	95	<5
× LV	95	10
× MD	0	45
× OC	95	45
× SW	60	50
× SC	0	55
× GB	90	55
× KV	55	55
× MJ	75	55
× CM	80	60
× RG	75	60
× LK	40	90
× MO	30	135
× MY	0	150
× SLO	15	180
× ORE	100?	300

square area, making for greater ease and accuracy of counting.

It was decided that since the pollen stainability for any one hybrid population might vary over a range of 5 to 20 per cent, the data should be evaluated within the nearest decile. Some evidence was obtained to indicate that pollen stainability is comparable with quality of seed set. Hybrids which were open pollinated by bees were systematically checked for seed set and size of silique. In most cases, those plants which had a high pollen stainability were also setting normal siliques with well-developed ovules, whereas those with low pollen stainability produced abnormal, small siliques and shrivelled ovules.

POLLEN FERTILITY AND TAXONOMIC STATUS

The data on pollen fertility of the interstrain hybrids, when grouped in different ways, reveal certain significant trends. One such trend served as the basis for the author's taxonomic treatment of the *glandulosus* complex (table 1). What is to follow is offered as evidence for this treatment. It will be recalled that Morrison (1941) conferred subspecific status on the taxa, *S. glandulosus "typicus," S. secundus, S. albidus, S. pulchellus,* and *S. niger*. In the present study, after hybridizing several strains of each of these taxa in a variety of combinations, it became apparent that certain strains were set apart from all others in their fertility reactions as hybrids. These genetically isolated strains were usually regionally distinct and corresponded with a subspecies recognized by Morrison. The extensive interfertility tests that led to the discovery of these genetically isolated segments of the *glandulosus* complex are summarized in a series of tables (tables 4 to 6) and diagrams (fig. 6a to d).

The most clear-cut example of genetic isolation is the population from Tiburon Peninsula (TB) which showed a uniform lack of fertility in crosses with any other strain (table 4). This edaphically and spatially isolated population has for many years been considered a distinct species, *S. niger* Greene, but was placed in *S. glandulosus* as a subspecies by Morrison. Twenty-two different hybrid combinations (including a few reciprocals) have been made crossing the TB strain with other strains from each of the regional and taxonomic groups within the complex. None of the pollen fertilities of these hybrids was over 50% and most of them were in the 0–10% range of fertility. In view of the correlation that later will be shown to exist between pollen fertility of hybrids and distance intervening between certain parental strains, it is of particular

interest to note that fertility is low in hybrids between TB and strains from stations only a few miles away. Thus the nearby Mount Tamalpais strains of subsp. *pulchellus* (BJ and RC), and of subsp. *secundus* (SG), as well as the BH strain of *S. albidus* from Alameda County are as strongly genetically isolated from TB as

TABLE 4. *Hybrid pollen fertility arranged according to taxa:* S. albidus, S. niger *and* S. glandulosus pulchellus × *all other strains*

Cross	No. of crosses with fertility percentages of					
	0–19	20–49	50–69	70–79	80–89	90–100
albidus × *glandulosus*	19	0	0	0	0	0
albidus × *secundus*	15	0	0	0	0	0
albidus × *pulchellus*	6	0	0	0	0	0
albidus × *niger*	3	0	0	0	0	0
albidus × *albidus* (& reciprocals)	0	0	3	0	0	9
Average fertility—*albidus* × *albidus* = 85%						
	0–9	10–19	20–29	29–50		
niger × *glandulosus*	6	5	0	0		
niger × *secundus*	5	0	1	0		
niger × *pulchellus*	1	0	0	0		
niger × *albidus*	1	2	0	1		
Total each percentile	15	7	1	1		
Proportion each percentile (%)	59%	32%	4.5%	4.5%		
	0–49	50–69	70–79	80–89	90–100	
pulchellus × Marin County (*secundus*)	0	0	0	0	6	
pulchellus × Sonoma County (*secundus*)	2	4	3	4	4	
pulchellus × North Interior	4	2	1	2	0	
pulchellus × *niger*	1	0	0	0	0	
pulchellus × *pulchellus*	0	0	0	0	1	
pulchellus × North Coast	1	2	2	0	0	
pulchellus × South Coast	3	0	0	0	0	
pulchellus × *albidus*	8	1	0	0	0	

TABLE 5. *Hybrid pollen fertility arranged according to taxa:* S. glandulosus *subsp.* secundus × *subsp.* secundus *and Oregon subsp.* secundus × *all other strains*

Cross	No. of crosses with fertility percentages of				
	0–49	50–69	70–79	80–89	90–100
secundus × *secundus**					
Sonoma County × Sonoma County	0	0	0	1	27
Marin County × Marin County	0	0	0	1	2
North Coast × Marin County	0	0	2	1	0
North Coast × Sonoma County	0	1	0	1	8
Marin County (LV) × Sonoma County	1	0	3	2	5
Marin County (SG) × Sonoma County	2	1	4	2	1
Oregon subsp. *secundus* × other strains					
Oregon × *secundus*	3	0	1	1	2
Oregon × *glandulosus*	0	3	3	3	0
Oregon × all others	3	0	0	0	0

* Includes North Coast *glandulosus*.

TABLE 6. *Hybrid pollen fertility arranged according to taxa:* S. glandulosus *subsp.* secundus × S. glandulosus *subsp.* glandulosus

Cross	No. of crosses with fertility percentages of				
	0–49	50–69	70–79	80–89	90–100
secundus × North Interior *glandulosus*	11	8	3	5	25
secundus × South Coast *glandulosus*	6	0	1	0	0
secundus × North Coast *glandulosus*	0	0	0	2	3

are those from much more distant localities. It seems justified on grounds of morphological distinctness, spatial isolation, and the evidence of genetic isolation to regard the TB strain as the distinct species, *S. niger,* just as did E. L. Greene many years ago.

Another group of strains that are set apart from all others on grounds of morphology, distribution, and interstrain fertility is the aggregation of populations occurring just south and east of San Francisco Bay and taxonomically identified here with *S. albidus.* The pollen fertility data for the *albidus* strains are given in table 4 and summarized in Figure 6a. It will be seen that while they are quite interfertile among themselves, the four strains of *albidus* are uniformly sterile in combination with any other strain. The inference to be drawn from this is then that *S. albidus* ranks with *S. niger* as a

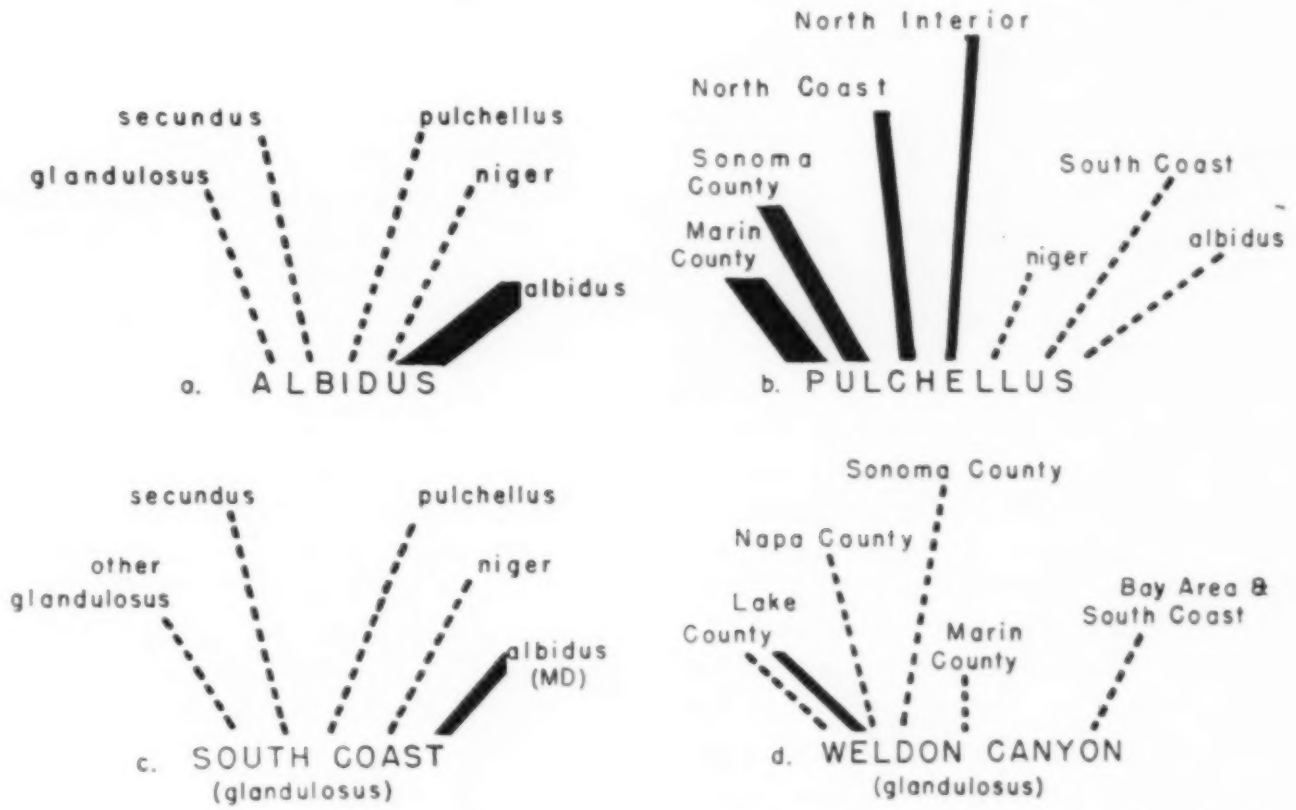

FIG. 6. Interfertility relationships involving certain members of the *Streptanthus glandulosus* complex.

Dotted lines indicate low interfertility; solid lines, high interfertility with widest solid lines indicating highest interfertility. North Coast, North Interior, South Coast, Napa County, and Lake County constitute strains of *S. glandulosus* subsp. *glandulosus;* Sonoma County and Marin County comprise strains of subsp. *secundus,* while Bay Area includes all strains of *S. albidus* and *S. niger.* 6a. *S. albidus* in combination with all other taxa. 6b. *S. glandulosus* subsp. *pulchellus* with all other taxa. 6c. South Coast (subsp. *glandulosus*) with all other taxa. 6d. Strain WC of subsp. *glandulosus* with other strains of subsp. *glandulosus* and with subsp. *secundus.*

well-defined species. It is unfortunate that more South Coast strains were not available for crossing, since the range of *S. albidus* often coincides with that of *S. glandulosus* subsp. *glandulosus* in certain areas south of San Francisco Bay. It is possible that the SLO strain which showed a pollen fertility of 25 per cent in crosses made both ways with the *albidus* strain, MD, should be placed in *S. albidus,* since it resembles more the lilac-flowered *albidus* populations (BH, MD, and SC) than the dark purple South Coast populations of subsp. *glandulosus.*

The three other taxa of the *Streptanthus glandulosus* complex are here accorded subspecific status. The evidence which has led to this disposition comes from three sources: Flower color and its regional homogeneity, the contiguous distribution patterns, and the high average pollen fertility among the twenty-six strains sampled from the three subspecies. Tables 4 to 6 and figures 6 and 7 show that most of the hybrid combinations made with various strains of the three subspecies *glandulosus, secundus,* and *pulchellus* gave pollen fertilities of over 50 per cent, and many of these as high as crosses within subspecies. Thus, similar flower color and geographic restriction tend to preserve the identity of each of the three taxa, while the high average fertility among them denotes the absence of a strong barrier to gene exchange. This latter criterion of high interpopulational fertility is a widely accepted one for subspecific rank and is invoked here.

Disposal of the majority of populations of the *glandulosus* complex either as species or subspecies poses no special problems. Within topographically circumscribed regions the subspecies are relatively uniform and interstrain fertility is high. A good example is the population system making up subsp. *puchellus,* which is morphologically homogeneous, is restricted to a small area in Marin County, and the strains sampled are fully interfertile as far as has been tested (table 4 and figure 6b).

Not all populations are as clearly delimited as those of subsp. *pulchellus.* The problem of subspecific homogeneity arises in the consideration of subsp. *secundus* and subsp. *glandulosus.* Apart from the anomalous strain from Josephine County, Oregon (ORE), subsp. *secundus* appears to be divisible into the Marin County populations and those from Sonoma County. The data on pollen fertility from *secundus* are summarized in table 5 and figure 7g–k. It will be seen that the two color morphs corresponding to the two geographic subdivisions of *secundus* tend to give lower pollen fertilities in combinations between the two groups than to hybrids within each group. The Oregon strain is placed in subsp. *secundus* only provisionally on the basis of similar flower color.

Turning to the more widespread and variable subsp. *glandulosus,* the principle of regional homogeneity still holds. Pollen fertilities of hybrids involving strains within the subsp. *glandulosus* group are given in table 7 and summarized in figure 7a–f. Strains constituting the North Coast and North Interior groups are with but very few exceptions highly interfertile. However, either of these two regional facies in crosses with the two *glandulosus* strains from the South Coast region yield only sterile hybrids. Although the strain, SLO, is somewhat set apart from other strains of subsp. *glandulosus,* the MY strain could just as well have come from any of the North Bay counties as from Monterey County, it is so similar morphologically to the North Interior

FIG. 7. Frequency distributions for pollen fertility of hybrids between strains belonging to subsp. *glandulosus* and subsp. *secundus.*

The ordinate begins at the zero abscissa for all graphs. 7a–f. Crosses within subsp. *glandulosus.* 7g–k. Crosses within subsp. *secundus.* 7l–n. Crosses between *secundus* and *glandulosus.* N. Int. = North Interior subsp. *glandulosus;* N. Co. = North Coast subsp. *glandulosus;* S. Co. = South Coast subsp. *glandulosus;* sec. = subsp. *secundus;* Son. = Sonoma County subsp. *secundus.*

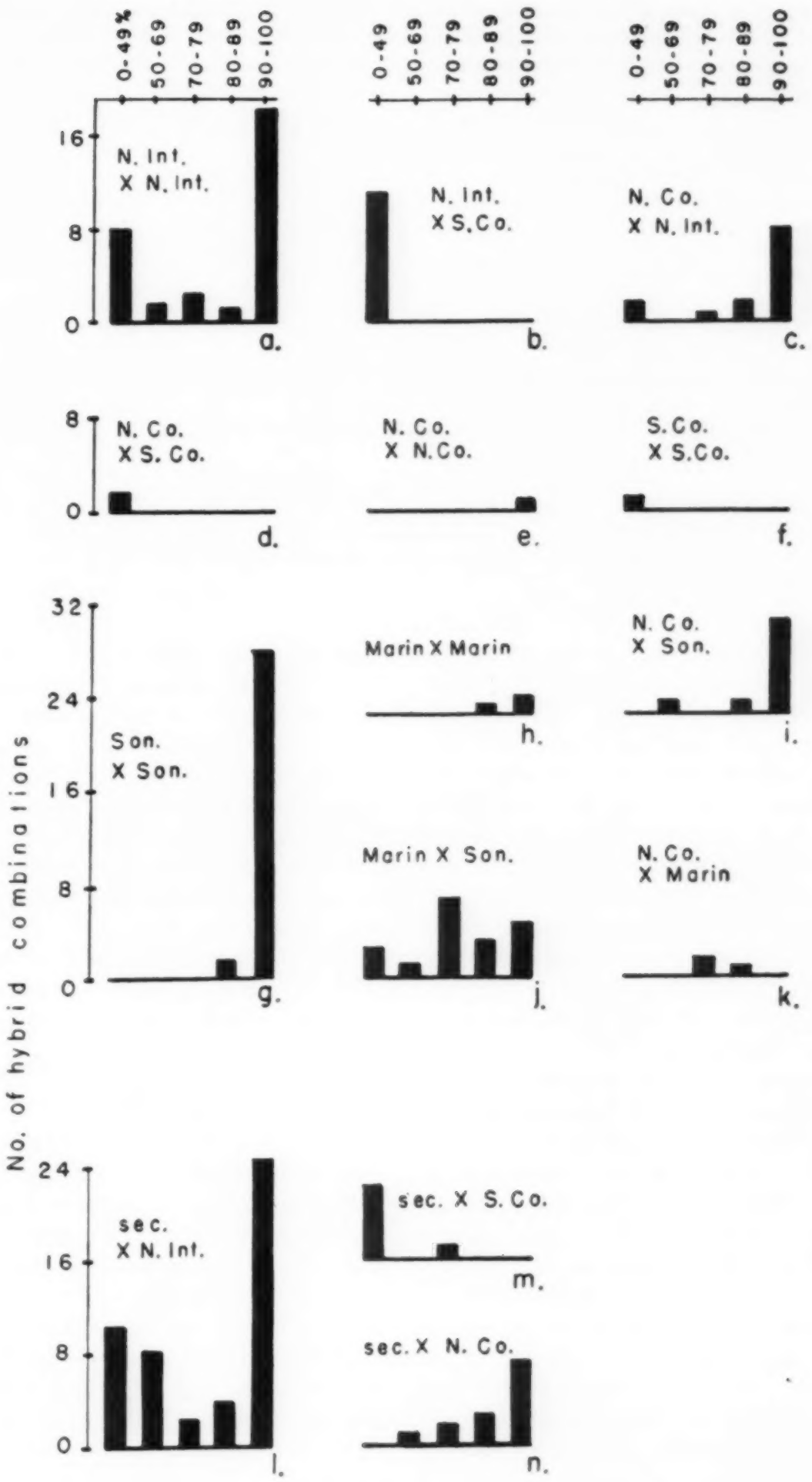
0-49%
50-69
70-79
80-89
90-100
16
8
0
N. Int.
X N. Int.
a.
0-49
50-69
70-79
80-89
90-100
N. Int.
X S. Co.
b.
0-49
50-69
70-79
80-89
90-100
N. Co.
X N. Int.
c.
8
0
N. Co.
X S. Co.
d.
N. Co.
X N. Co.
e.
S. Co.
X S. Co.
f.
32
24
16
8
0
No. of hybrid combinations
Son.
X Son.
g.
Marin X Marin
h.
N. Co.
X Son.
i.
Marin X Son.
j.
N. Co.
X Marin
k.
24
16
8
0
sec.
X N. Int.
l.
sec. X S. Co.
m.
sec. X N. Co.
n.

FIG. 7.

TABLE 7. *Hybrid pollen fertility arranged according to taxa:* S. glandulosus *subsp.* glandulosus × *subsp.* glandulosus, *WC* × *other strains, and South Coast* × *other strains*

	No. of crosses with fertility percentages of				
	0–49	50–69	70–79	80–89	90–100
North Interior × North Interior	8	2	3	1	19
North Interior × South Coast	12	0	0	0	0
North Coast × North Interior	2	0	1	4	8
North Coast × South Coast	2	0	0	0	0
North Coast × North Coast	0	0	0	0	1
WC × Lake County *glandulosus*	2	0	0	0	1?
WC × Napa County *glandulosus*	1	0	0	0	0
WC × Sonoma County *secundus* & *glandulosus*	3	0	0	0	0
WC × *niger*	1	0	0	0	0
WC × Bay Area & South Coast	3	0	0	0	0
	0–10	11–20	21–30		
South Coast × other strains	27	2	2*		
South Coast × South Coast	1	0	0		

* SLO × MD & reciprocal.

group. Herbarium specimens of plants from the several South Coast Range counties compare closely with North Coast and North Interior plants. Though only two samples from these southern localities have been tested for interstrain fertility, it is suspected that the others from south of San Francisco Bay would show low pollen fertilities with both *S. albidus* and with subspecies *glandulosus* from north of the Bay. Apparently the South Coast strains achieved genetic isolation in advance of any significant morphological divergence.

The behavior of the Weldon Canyon (WC) strain of North Interior *glandulosus* has been singled out in table 7 and figure 6d. Although somewhat darker purple in flower than some other strains of subsp. *glandulosus,* WC plants closely resemble all other strains of this subspecies. Yet in its fertility reaction with all other strains, it is almost as sterile as crosses involving *S. niger* (TB). Geographically, the WC strain is perhaps more isolated than most others of subspecies *glandulosus.* It occurs in the Vaca Mountains of western Solano County, one of the easternmost stations for any member of the Pulchelli section. That the cross, KX × WC, is actually a fertile hybrid is open to question. The two parents are so similar that the fertile plants could just as well be accidental maternally selfed progeny (i.e., KX F_1s). That this is a likelihood is supported by the much lower fertility (0 per cent) for the hybrid WC × PO, in which the male parent was collected 25 miles closer to WC than was KX. In summary, it would seem that the WC strain is another instance of genetic isolation having preceded morphological divergence.

VARIATION OF POLLEN FERTILITY WITHIN AND BETWEEN TAXA

It has been emphasized in the previous section that high pollen fertility of hybrids between strains of similar flower color and from the same geographic province is the cohesive element that binds populations of Streptanthus into taxa of varying levels. Yet it was also pointed out that pollen fertility of some interstrain hybrids within these taxa may vary, often significantly. An intriguing correlation that often transgresses taxonomic boundaries has come to light upon closer examination of this variability in pollen fertility. It appears that *the level of hybrid pollen fer-*

tility varies inversely with the distance in miles separating the strains which were crossed. Often, topographic barriers may accentuate the effect of the intervening distance between strains.

A clear-cut example of this "fertility/distance correlation" is seen in the data for crosses within subsp. *secundus* (tables 3 and 5, and fig. 8c). Thus there is a significant reduction in fertility of hybrids

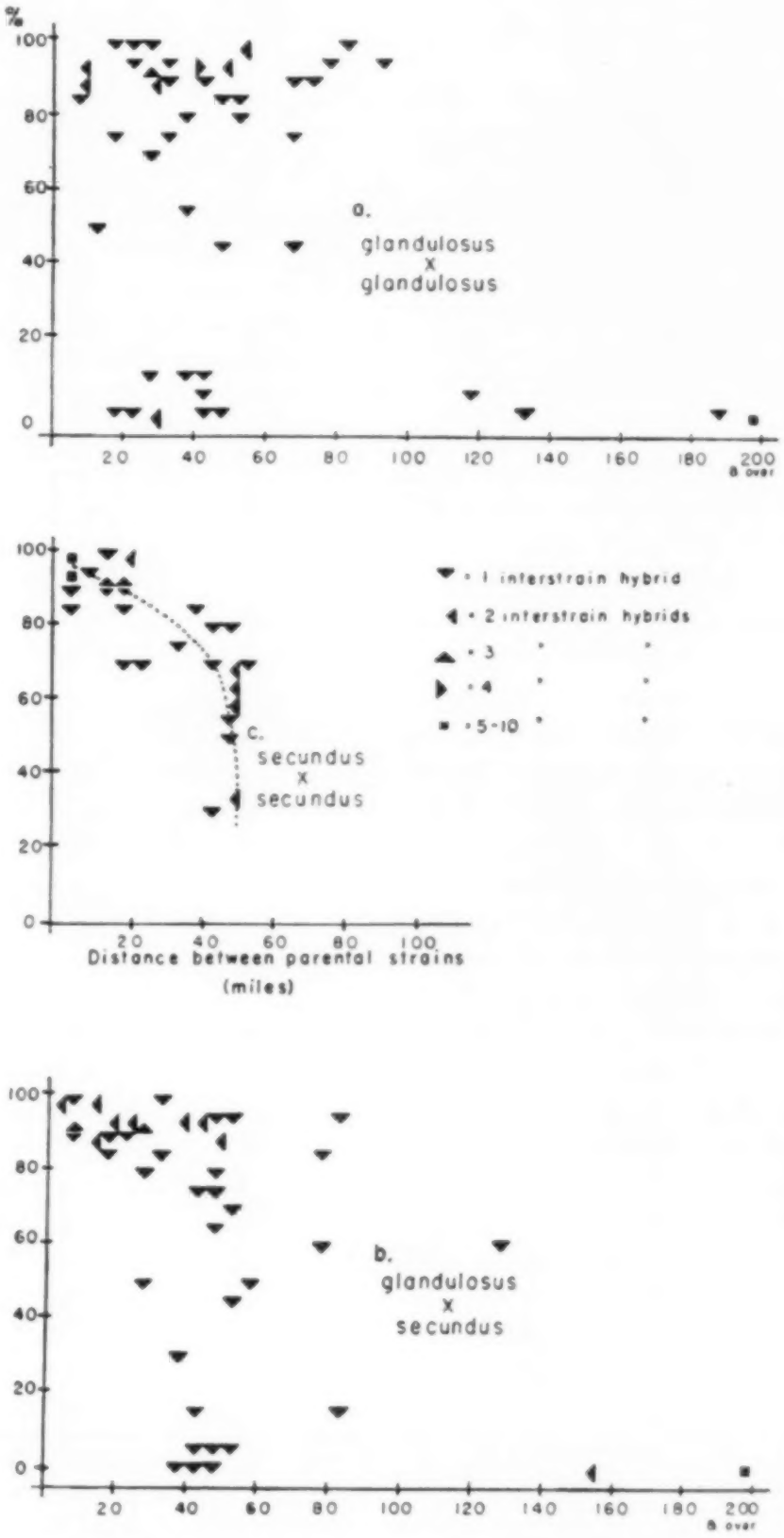

FIG. 8. Relationship between pollen fertility (vertical axis) and distance between parental strains for crosses of strains within and between subsp. *glandulosus* and subsp. *secundus*.

The triangular and square symbols indicate the number of different interstrain hybrids at a given coordinate. The coefficient of correlation (r) and r^2 for figs. 8a–c are given in the text.

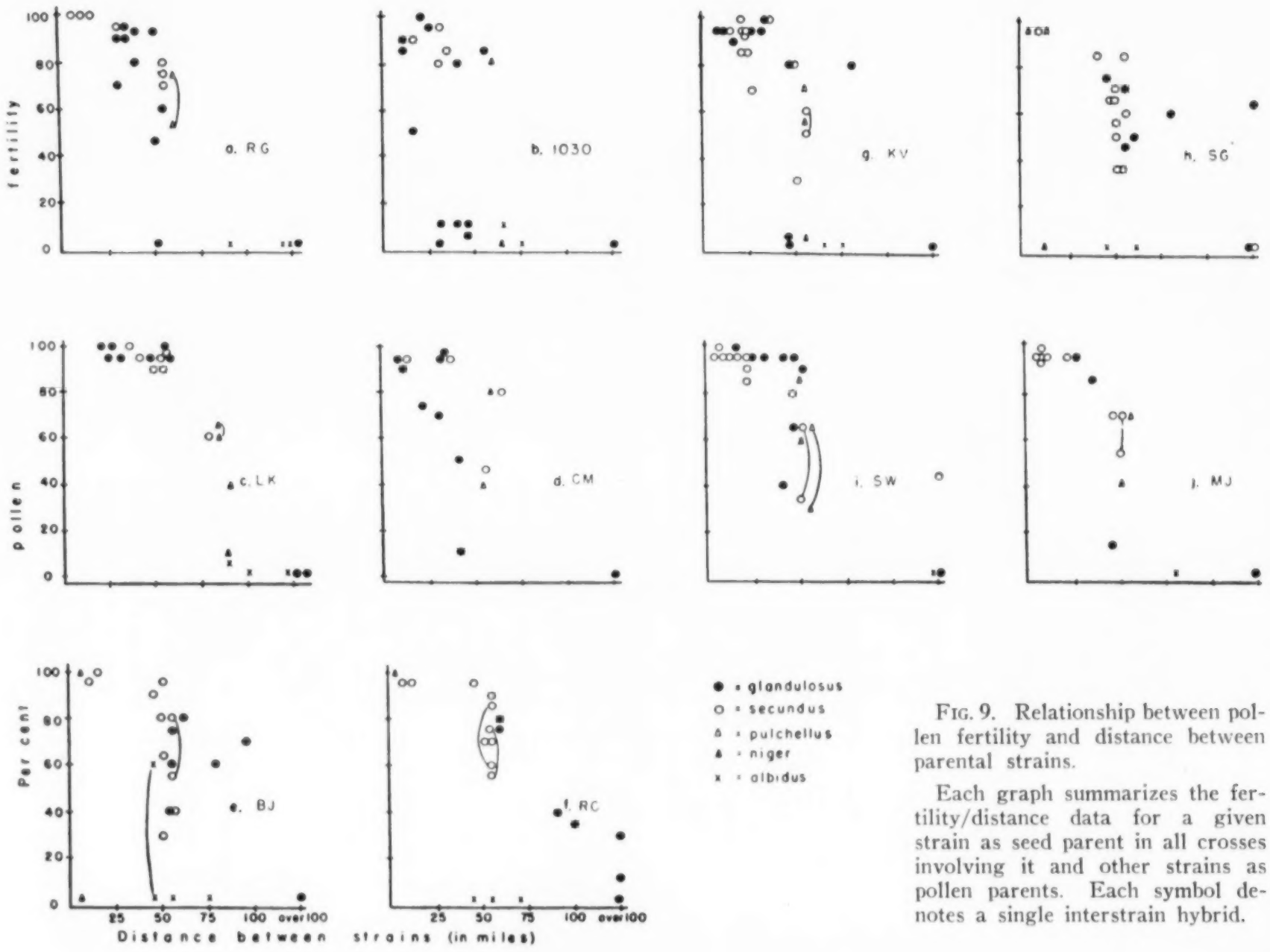

FIG. 9. Relationship between pollen fertility and distance between parental strains.

Each graph summarizes the fertility/distance data for a given strain as seed parent in all crosses involving it and other strains as pollen parents. Each symbol denotes a single interstrain hybrid.

between Marin and Sonoma county strains in comparison with Marin × Marin and Sonoma × Sonoma hybrids. No apparent topographic barriers exist between the two groups of populations, since available habitats occur in a partially discontinuous pattern from Mount Tamalpais in Marin County to north of Guerneville in Sonoma County. A similar, though less well-defined trend holds for the hybrids of strains in subsp. *glandulosus* (tables 3 and 4, and fig. 8a). The farther apart that two strains of subsp. *glandulosus* may occur in the wild, the lower the fertility of their hybrid may be. The data for pollen fertility of hybrids between subsp. *glandulosus* and subsp. *secundus* in various interstrain combinations is plotted against distance in figure 8b. Again, the trend of reduction in fertility as a function of increasing distance between strains is evident.

The two statistics, coefficient of correlation (r) and r^2, have been calculated for the three sets of data illustrated in figure 8. For figure 8a, $r = -0.64$ and $r^2 = 0.41$ (d.f. = 67); for figure 8b, $r = -0.70$ and $r^2 = 0.49$ (d.f. = 59); for figure 8c, $r = -0.81$ and $r^2 = 0.64$ (d.f. = 46). As would be suspected simply by inspection of the graphs, the correlation is statistically highest for the *secundus* × *secundus* crosses, although the other two sets of data show a statistically significant correlation. The calculation of r^2 (Snedecor, 1946, p. 157) is intended to bring out the partitioning of correlation due to the interaction of fertility and distance on the one hand (r^2) and the correlation which is independent of distance on the other hand ($1 - r^2$).

The "fertility/distance correlation" is again apparent when the data on fertility of hybrids are analyzed without regard for subspecific boundaries. In a number of instances, members of different subspecies that inhabit adjacent territory are often more interfertile than are those strains of the same subspecies where spatial separation is much greater. For example, many North Interior strains of

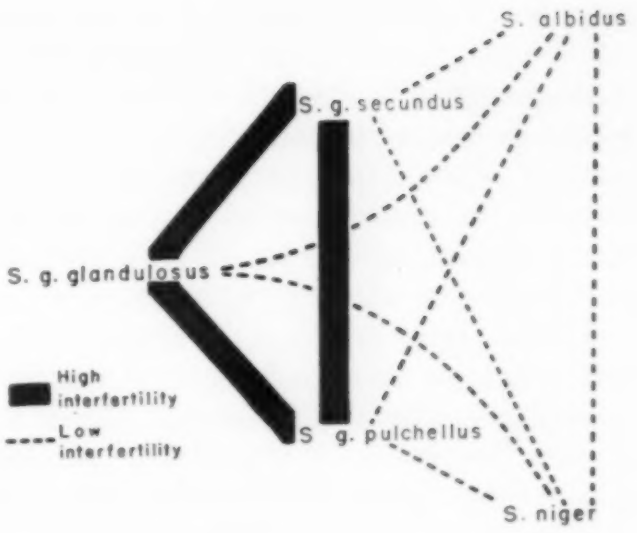

FIG. 10. Summary chart of the interfertility relationships among the five taxa of the *Streptanthus glandulosus* complex.

subsp. *glandulosus* are more fertile in hybrids with subsp. *secundus* from Sonoma County than they are with other strains of *glandulosus* (table 3; also compare fig. 7l with figs. 7a, c). The same comparison can be made for Marin County subsp. *secundus* in crosses with Marin County subsp. *pulchellus* versus crosses involving Marin County subsp. *secundus* and Sonoma County subsp. *secundus* (tables 4, 5 and fig. 6b).

An equally instructive if not wholly comparable example is provided by the hybrids with the Russian Gulch (RG) strain from coastal Sonoma County (table 3 and fig. 9a). On the grounds of flower color (rose-lilac), this strain is intermediate between subsp. *glandulosus* and subsp. *secundus* and hence was rather arbitrarily placed in subsp. *glandulosus*. It is equally fertile with certain strains of both subspecies, showing pollen fertilities of over 90 per cent with four strains of subsp. *glandulosus* and two strains of subsp. *secundus* (table 3). These six strains are all within 50 miles of RG and in a region of no sharp habitat discontinuity. Other strains of both subspecies occurring at greater distances from RG give lower pollen fertilities in hybrids. This additional example of the fertility/distance correlation suggests that the regional homogeneity mentioned earlier may

exist only negatively as a lack of any barrier to gene flow between morphologically distinguishable strains, all of which are in spatial proximity to one another. In such cases of regional sympatry and high interfertility, the designation of strains as elements of one subspecies or the other is purely on a morphological basis.

The sharpest break in the range of hybrid fertilities occurs where strains originating from the San Francisco Bay area or the South Coast region are crossed with strains from the counties north of the Bay. No matter whether the crosses are at the inter- or infraspecific level, the hybrids are invariably infertile (see tables 4–7 and figs. 6c and 7b, f, m. It cannot be proved directly that San Francisco Bay, *per se,* constitutes the barrier to gene exchange and hence a basis for genetic isolation. However two facts are of interest with regard to this speculation. First, the Bay interposes a land break in the Jurassic Franciscan formation; this geological formation contains the rock types from which are derived the soils inhabited by members of the *Streptanthus glandulosus* complex. East of the Bay, *S. glandulosus* would encounter Cenozoic sediments, combined with an unsuitable topography and associated vegetation. The closest Jurassic Franciscan rocks north and south of the Bay are separated by the Golden Gate! The second point is the remarkably comparable situation in Godetia (= Clarkia, Lewis and Lewis, 1955), discovered by Hiorth (1942) and summarized by Stebbins (1950). Following the nomenclature of Lewis and Lewis, members of the *Clarkia amoena-rubicunda* complex extend along the Pacific Coast from British Columbia to counties just south of San Francisco Bay. As with the *S. glandulosus* complex, the Bay coincides with a fertility barrier for Clarkia. *Clarkia amoena,* north of the Bay is interfertile throughout its range as is *C. rubicunda* south of the Bay. But *C. amoena* × *C. rubicunda* crosses are sterile. Whether or not the Bay itself may have initiated the development of a barrier to crossing, the coincidence between the Bay as a topographic gap in plant distribution and the reduction in fertility of crosses between strains north and south of the Bay certainly suggests a relationship between the two barriers.[1]

The fertility reactions of each of several strains with all those serving as opposing parents will now be examined in the light of the fertility/ distance correlation. Fertility data for crosses between a given seed parent and all strains used as pollen parents have been taken from table 3 and plotted as a function of the distance intervening between parent strains (fig. 9). All combinations with a given seed parent are plotted without regard to the taxonomic affinity of the paired strains. Reciprocal combinations of the same cross are shown as connected lines between the reciprocals. In general, as distance in miles between parental strains increases, fertility in the hybrids between them is reduced, and usually precipitously after 50–75 miles of intervening distance. The drop in hybrid fertility occurs in relation to at least three sets of conditions: (1) When the intervening distance between parental strains involves some topographic or ecologic barrier (e.g., San Francisco Bay, mountain ranges, alluvial soils of major valley systems, or absence of suitable habitats in otherwise topographically likely areas). (2) Considerable intervening distances without habitat discontinuity (e.g., Marin versus Sonoma County subsp. *secundus,* and Mendocino County versus Napa County subsp. *glandulosus*). (3) Small (minimal) intervening distances with no apparent extrinsic (environmental) barriers to gene exchange (e.g., *S. niger*-TB × Mount Tamalpais *pulchellus* and *se-*

[1] An example which suggests that spatial separation by the Bay need not be correlated with genetic isolation is offered by Clausen *et al.* (1940). Two races of the maritime ecotype of *Layia platyglossa* which are separated by the Golden Gate and by 150 miles formed fertile hybrids and their F_2 progeny showed no segregation for the characters of the ecotype.

cundus; South Coast *glandulosus* MY × SLO, and others). This third condition has been discussed in connection with the genetic basis for delimitation of the taxa, *S. albidus* and *S. niger* from *S. glandulosus.*

Meiosis in the Hybrids

It was pointed out that the haploid chromosome number for the *Streptanthus gladulosus* complex is 14, and that all strains thus far sampled have shown normal meiosis. Because of the marked reduction in fertility in many of the hybrids, it was desirable to determine whether the infertility could be traced to some meiotic irregularity. Accordingly, fifty-five of the hybrids, ranging from 90 per cent to 1 per cent stainable pollen were examined for possible irregularities in chromosome behavior during and after meiosis of pollen mother cells. Neither the hybrids with high nor low pollen fertility showed any significant abnormal behavior in microsporogenesis. At diakinesis, 14 pairs were invariably present. Occasionally, what appeared to be lagging univalents were seen at first and second anaphases, but these were never found to be excluded from first or second telophase nuclei. Micronuclei and other post-meiotic signs of irregularity were not found, pollen quartets thus having a completely normal appearance.

It was hoped that by a cytological examination of stages from diakinesis to mature pollen, the stage could be discovered at which the apparently normal meiotic or post-meiotic behavior gave way to the production of inviable pollen. To ascertain this, the buds from several inflorescences were examined consecutively from youngest to oldest, but without success. The search for a cytological basis for this breakdown in microsporogenesis is continuing.

From these observations, it seems that there are no visible features of microsporogenesis that can account for the production of inviable pollen in hybrids of reduced fertility.

Discussion

Taxonomic Evaluation

Like the morphological criteria of classical taxonomy, the results of experimental studies in taxonomy must be evaluated if they are to serve in any taxonomic revision of merit. Whether or not data from breeding studies can be used to achieve a taxonomic end will often depend upon the purely practical goals of plant classification. Thus many biosystematic investigations must seek a compromise with classical taxonomy in order to present a workable taxonomic treatment of a group. Results from the present study lead to a taxonomic interpretation of the *Streptanthus glandulosus* complex that synthesizes several elements of the treatments by Jepson (1925), Morrison (1941) and Abrams (1944). Figure 10 summarizes the relationship and taxonomic status of the five taxa as dictated primarily by the results of interfertility tests. Three of the five taxa (*S. glandulosus, S. albidus,* and *S. niger*) are regarded as species. Heretofore, Jepson, Morrison, and Abrams had regarded *S. albidus* as an infraspecific element of *S. glandulosus;* Morrison, however, was the only one of the three to reduce *S. niger* to the subspecific level. *S. glandulosus* is defined here as a rather polymorphic species of wide range in California; it consists of three subspecies with more or less well-delimited morphological and geographic ranges, and with high average inter- and infrasubspecific fertilities. The three subspecies, *S. glandulosus* subsp. *glandulosus,* subsp. *pulchellus,* and subsp. *secundus* correspond to three of the five subspecies proposed by Morrison (1941).

The exceptional infertility of certain hybrids between strains from within as well as between subsp. *glandulosus* and subsp. *secundus* points to the essential disagreement that often exists between evidence from morphology and from hybridization. It will be recalled from the earlier section on Taxonomy that some populations of subsp. *glandulosus* were

wholly infertile with others in the same subspecies or with any other strains in the complex. For example, the strains from Weldon Canyon (WC) in Solano County and from Nacimiento Creek (MY) in Monterey County formed sterile hybrids with all other strains, even with those strains morphologically indistinguishable from WC and MY. Yet other strains of subsp. *glandulosus* were highly interfertile and as well formed fertile hybrids with strains of the morphologically distinctive subspecies, subsp. *secundus* and subsp. *pulchellus*. Stebbins (1950) has analyzed the problem of correlation that may exist between degree of morphological resemblance and degree of interfertility as it relates to making taxonomic judgments. Of the five examples analyzed by Stebbins for such correlation, three appeared to show no correlation and two showed correlation. He concludes that when the genetic factors for morphological features and for isolating mechanisms are linked, correlation between the two sets of factors may exist. However, as often as not, the necessary genetic linkage either may not occur or may be rather easily broken by crossing over within the linkage group. From the present study it is concluded that the correlation between morphological resemblance and genetic compatibility may be high or low. Also, the presence or absence of the correlation appears to be conditioned by the spatial distance separating the parental strains of the hybrid.

Despite the development in recent years of a rich literature on many aspects of experimental taxonomy, relatively few investigations have been concerned with testing the fertility of hybrids between strains of the same species complex. In connection with the results presented in this paper, reference can be made to the work of Snyder (1950, 1951) on interpopulational fertility in the grass species, *Elymus glaucus* Buckl. Snyder analysed 34 hybrid combinations involving twenty strains taken along an altitudinal transect of the central Sierra Nevada of California. Crosses of some strains yielded fully fertile hybrids; others gave marked reduction in fertility. Although the 20 strains could be placed tentatively in 15 ecospecies on the basis of hybrid compatibilities, evidence from morphological, cytological, and breeding studies favored the retention of the variants within the cenospecies, *E. glaucus*. Snyder's disposition of his twenty strains of *E. glaucus* can be compared with the placement of 27 strains of *Streptanthus glandulosus* (as here delimited) into the three subspecies mentioned above. In both Elymus and Streptanthus, morphological resemblance has taken precedence over the interfertility results, which by themselves would dictate the recognition as species of those strains that are genetically isolated. This action is justified on the grounds that such strains are not totally isolated (as is the case of *S. albidus* and *S. niger*) but are interfertile with other strains within the same morphological spectrum of affinity.

Hybrid Cytology

The absence of any detectable meiotic irregularity in even the most highly sterile of the Streptanthus hybrids is as yet unexplained. Nor, as was pointed out, is it possible to discover the stage of microsporogenesis at which the breakdown leading to inviable pollen occurs. It is easy to generalize that the pollen sterility results from chromosomal or genic disharmonies in the hybrids. But securing proof for one or the other of these causes of sterility is not so easy!

In recent years similar cases of sterility without well-defined meiotic irregularities have been reported (see Stebbins, pp. 221–222, 1950, for extensive literature up to 1950). More recently a particularly comparable situation to the one in Streptanthus was reported by Lewis (1953). Analyzing certain crosses between pairs of individuals from different populations of both *Clarkia dudleyana* and *C. biloba*, Lewis found that the hybrid sterility was not associated with any detectable meiotic irregularity. Despite the absence of

meiotic irregularity, most investigators have proceeded to interpret the hybrid sterility as either genic or chromosomal, or both. In dealing with this problem, Stebbins (1950) hypothesizes that while chromosomal differences leading to structural heterozygosity in hybrids do exist in many of the examples he cites, the differences may be so small as not to interfere with the meiotic process. To this interpretation of infertility despite normal meiosis, Stebbins (1945) gave the name "cryptic structural hybridity." Two lines of evidence are adduced to support the hypothesis: (1) It should be possible to remove (or reduce) the sterility imposed by structural heterozygosity by doubling the chromosome number of the hybrid. (2) Often the structural hybridity may not be wholly "cryptic," and occasionally meiotic disturbances such as bridge-fragment configurations at anaphase, lagging univalents, etc., will be visible. The former test has not been tried for Streptanthus, nor has evidence of the latter type been detected for certain thus far. It seems wiser, therefore, to label the sterility simply as "cryptic sterility" in the present case until more can be learned about meiosis in relation to sterility in hybrids of Streptanthus.

Relationship Between Interfertility and Isolation

The detailed analysis of interstrain fertility in the *Streptanthus glandulosus* complex has led to the discovery of a statistically significant inverse correlation between the degree of fertility of hybrids and the distance in miles separating the two parents. This correlation, documented in an earlier section, can be summarized as follows: The level of fertility of hybrids between strains belonging to the three subspecies of *S. glandulosus* Hook. tends to decrease as the distance between the strains serving as seed and pollen parents increases. From figure 8 it was learned that the correlation held for the various interstrain hybrids both within and between the two subspecies, subsp. *secundus* and subsp. *glandulosus*. Figure 9 presented ten separate fertility/distance correlations for interstrain hybrids involving ten different strains as seed parents, each crossed with strains from all of the taxa in the complex. Not only do these results document a significant trend in relationship between pollen fertility and distance, but in particular, figure 8b and figure 9 illustrate the fact that this trend often transgresses subspecific limits.

One feature of these graphs merits additional comment here. The clustering of some of the fertility/distance plots in the low fertility/small intervening distance sectors of graphs (figs. 8a and 8b) suggests that genetic isolation may develop between certain strains separated by short distances. It is also possible that these low fertility/short distance points on the graphs are manifestations of some environmental barrier (topography, soil, etc.) which cannot be dealt with independently of the distance factor.

There seems to be little precedent in the literature on isolating mechanisms which could aid in interpreting the fertility/distance correlation. In most instances where intraspecific hybrids have been studied, either the hybrid fertility has been high, or if any variation in hybrid fertility was noted, it did not exhibit any relation to the intervening distance between parental strains. An example of this second alternative—absence of a relationship between variability in hybrid fertility and geographic origin of parental strains—appears in the work of Stebbins and Vaarama (1954). Summarizing crossability relations among a number of strains of the two related grass species, *Elymus glaucus* and *Sitanion jubatum*, the authors point out that ease of crossing between the two groups of strains bore no obvious relationship to the geographic origin of the strains. The studies of two other experimental taxonomists, Dr. Verne Grant and Dr. Harlan Lewis, bear more directly on this discussion of the fertility/distance correlation, since, in the

course of their research, both workers independently have obtained evidence for a relationship between fertility and distance.

In two groups of the genus Gilia (Grant, 1952b; 1954b), the *G. tricolor* complex as well as other leafy-stemmed gilias, interstrain hybrids were usually readily obtained and the plants were vigorous and fertile, regardless of the distance between strains. However, the results of experimental hybridization in the *Gilia achillaefolia* complex (Grant, 1954a) disclosed the presence "of at least three compatibility groups, a northern, a southern, and a central group." With but a single exception, crossing attempts between northern and southern strains were unsuccessful. This, of course, is an example of an even more stringent barrier to gene exchange than that found in Streptanthus where hybridizations are invariably successful even though the F_1s may be infertile. Grant interprets this interpopulational incompatibility as "the result of some fairly simple genic reaction and . . . the determining gene . . . represented in the different populations by different alleles." Still another example from Gilia is instructive (Grant, 1952a): Certain sympatric subspecies of *Gilia capitata* are readily crossed, while distantly allopatric ones are not. For example, the cross *G. capitata* subsp. *abrotanifolia* (from Kaweah) × *G. capitata* subsp. *pedemontana* (from Kaweah) is easily made and the F_1s quite fertile, whereas the cross *G. capitata abrotanifolia* (from Mentone) × *G. capitata pedemontana* (from Kaweah) is obtained with difficulty. Mentone in the San Bernardino Mountains is roughly 190 miles south of Kaweah in the foothills of the Sierra Nevada. Grant's discovery of these differences in interpopulational fertility led him to conclude that "These results definitely run counter to expectation on the hypothesis that the sympatric strains ought to show the greatest degree of incompatibility. On the contrary, if there is any correlation to be found in these data, it is that *compatibility decreases in proportion to geographic remoteness.*"

Two examples from the work of Harlan Lewis in the genus Clarkia are likewise relevant to this discussion. Lewis (1953) made 200 interpopulational crosses of *C. deflexa* involving 30 combinations of 11 different populations. Some were fully fertile, while others produced F_1 hybrids that were sterile or in which the derived F_2 plants were largely degenerate or sterile. Although no direct correlation between geographic distribution and fertility was apparent, the combined effect of spatial and topographic isolation was noted. In considering the joint effects of distance and topography on genetic isolation, Lewis emphasizes a point that was brought out in connection with the interpopulational hybrids of Streptanthus. He states that "a two-dimensional representation of geographic relationships may be misleading." Thus two intersterile populations, though separated by a distance of about five miles were separated by at least 2000 feet in elevation and grew on opposite sides of a ridge. The barrier of San Francisco Bay, interposed between spatially adjacent populations of Streptanthus appears to be a comparable situation. In the same paper, Lewis discussed other cases of interpopulational sterility in Clarkia. A southern Californian race of *C. Dudleyana* crossed with one from the Sierra Nevada produced a sterile hybrid; certain populations of *C. biloba* were also intersterile. In both cases meiosis was normal.

In a more recent paper, Roberts and Lewis (1955) have elaborated on the *Clarkia biloba* example. Three subspecies, each composed of small, semi-isolated colonies, replace each other geographically along a north-south axis in the foothills of the Sierra Nevada. The two southern subspecies are completely interfertile while fertility is reduced to about 50 per cent in hybrids between the northernmost subspecies (subsp. *brandegeii*) and either of the two southern ones. The intersterility,

coupled with the abrupt morphological transition between the northern and southern races do not appear to have any adaptive value (i.e., any selective advantage for a particular habitat). Rather, the authors prefer to interpret the isolation as primarily geographic, which has resulted in the development of a factor which reduces fertility in certain heterozygotes (i.e., northern × southern subspecies).

Three significant parallels appear when the examples just cited from Gilia and Clarkia are compared with the present account of interpopulational compatibility in Streptanthus. The species in question from all three genera are (1) annuals with (2) similar ecological preferences (all three often occur together on serpentine), and (3) occur in small, semi-isolated populations. Combining these facts with two lines of experimental evidence from studies with the three genera —(1) that adjacent populations are usually interfertile and (2) that less adjacent ones are often intersterile—lends support to a hypothesis for the origin of interpopulational sterility: When present, the barrier of genetic isolation between strains may not have arisen under the pressure of selection to impair gene flow between contiguous populations. Rather, as Roberts and Lewis (1955) hypothesized for *Clarkia biloba,* it is possible that the divergence in morphological character and the development of genetic isolation are simply two independent consequences, either or both of which could result from non-adaptive, genetic divergence following geographic isolation.

It is acknowledged that the erection of a barrier to gene exchange between strains of *S. glandulosus* need not be solely non-adaptive in origin. Recently, evolutionists have been reluctant to invoke random fixation as the sole explanation for situations comparable to this one in Streptanthus. Until the nature of genetic segregation for factors of fertility and morphology in F_2 cultures is known and until the effect of selection is ruled out, it is perhaps wisest to leave the door open for a choice of either random fixation, selection, or a combination of both as final interpretations.

One of the most intriguing problems of serpentine endemism in Streptanthus is that of accounting for the origin of the disjunct distribution in many of the species. A direct solution to the problem is not likely to be found. However the fertility/distance trend as documented here for *S. glandulosus* offers indirectly a possible explanation. Since adjacent strains are often highly interfertile, they may represent populations which have not been spatially isolated for as long a time (or as effectively in space) as those separated by greater intervening distances or by topographic barriers. It is therefore suggested that populations of *S. glandulosus* may have been more confluent in the past than they are now. The present discontinuous distribution and restriction to serpentine may be the result of progressive elimination of biotypes adapted to non-serpentine areas. Evidence from competition experiments has been presented elsewhere (Kruckeberg 1954) supporting the contention that severe competition on the adjacent non-serpentine sites could have exerted a depleting effect on biotypes in the past. This interpretation of the origin of the present distribution pattern of *S. glandulosus* fits the pattern of endemic species described by Stebbins (1942) as a *depleted species*—". . . those which were formerly widespread and genetically diverse but have lost many or most of their biotypes." As was pointed out by the author (Kruckeberg, 1951), this explanation for the origin of endemism on serpentine is undoubtedly only part of the story. If the hypothesis of biotype depletion is invoked, then subsequent genetic fixation of population-specific morphological characters as well as of genetic isolating barriers must be postulated to have been later events leading to the present pattern. Furthermore, other serpentine endemics could have originated in other ways, fol-

lowing the pattern of *insular species* as described by Stebbins (1942). Since unequivocal proof of past origins of species is usually unattainable, evidence from indirect sources such as those examined here are at least helpful in developing working hypotheses.

Summary

Populations of the annual crucifer, *Streptanthus glandulosus* Hook., are largely restricted to the discontinuous outcrops of serpentine soil in California. Many of the partially isolated serpentine habitats support morphologically distinguishable strains of the species complex. Such polymorphism has led taxonomists in the past to recognize no less than 12 species. More conservative treatments, however, define the complex taxonomically as one or two species with several infraspecific taxa.

It is proposed that the spatial isolation imposed by the discontinuity of edaphically suitable habitats has led to the origin of the morphologically distinguishable strains of varying degrees of genetic isolation. To determine the extent of genetic isolation among the various strains, a program of hybridization involving 32 different strains of the *S. glandulosus* complex in over 300 combinations was undertaken.

Significant differences were encountered in the pollen fertility of the interstrain hybrids. These differences in fertility were used as a measure of genetic isolation. The fertility data also provided a basis for making taxonomic interpretations of the group. The complex was found to consist of three genetically isolated elements, which could be recognized morphologically as the species, *S. albidus* Greene, *S. glandulosus* Hook., and *S. niger* Greene. Of these, *S. glandulosus* is the most widespread and the most polymorphic in morphology and in range of interfertility. The distribution pattern of the morphological variants and the relative interfertility of the strains permitted the recognition of three subspecies within *S. glandulosus*.

The study of microsporogenesis reveals no significant meiotic irregularity in even the most highly sterile hybrids. Moreover it was not possible to detect the critical stage at which the pollen became defective. No evidence is yet available which would permit a decision on whether the sterility is genic or is determined by cryptic structural differences.

The analysis of pollen fertility of the artificial hybrids disclosed a significant inverse correlation between degree of hybrid fertility and the distance in miles separating the two parents. In other words, hybrid fertility tended to decrease as the distance between parental strains increased. This trend often transgressed subspecific boundaries. As yet it cannot be conclusively determined whether the barrier to gene exchange between certain populations is the result of random fixation, selection, or a combination of these two evolutionary mechanisms.

The fact that spatially adjacent strains of a given subspecies of *S. glandulosus* are more highly interfertile than those separated by greater distances leads to the hypothesis that the present distribution pattern has resulted from biotype depletion. Spatially adjacent biotypes are still interfertile, but those more completely isolated by distance have become intersterile following depletion of intervening biotypes.

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ADAPTIVE EVOLUTION IN THE AVIAN WING

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Introduction

The form of a bird's wing is so basically important to the successful exploitation of an ecological niche that it inevitably yields many instructive examples of adaptive evolution. It also provides interesting examples of convergence, as is to be expected of a structure that contributes materially to such important functions as locomotion and the obtaining of food. It may therefore be profitable to examine the principal categories of wing type (using the word aerodynamically rather than anatomically) and to see how they correlate with special requirements.

Space does not warrant a full discussion of the function of an airfoil, which is readily obtainable in adequate detail from elementary treatments of aircraft aerodynamics, such as those issued for pilot training; but attention must be drawn to some points that are vital to an appreciation of the various adaptions with which we are concerned.

Lift Devices

Most birds' wings are airfoils of high camber, adapted to provide high lift economically at low speeds. In section they resemble many early aircraft wings that were designed for operation at about 100 m.p.h. or less. As an airfoil moves through the air some lift is produced by positive pressure on the under side; but most of it, usually two-thirds or more, is due to negative pressure on the upper surface; the pressure distribution being somewhat as shown in figure 1. This negative pressure results from the positively curved upper surface functioning as a venturi. The increased velocity of the air passing over this curved surface causes a drop in pressure.

By increasing the angle of attack (fig. 1) lift can be increased up to a point at the cost of some extra drag, the latter being undesirable under most circumstances but useful during landing. As the angle of stall is approached, however, the airflow breaks away from the upper surface in turbulent eddies and lift quickly drops to a small fraction of the maximum. Lift can be increased and stalling delayed in several ways, the most important of which is the wing slot. Slots, which were frequently used in aircraft when speed ranges were low, are flattened venturis, figure 2 representing a typical section. A high-speed jet of air drives through the slot and follows the contour of the top of the wing, decreasing pressure and also delaying the turbulent break-away of airflow that constitutes the stall. Two types of wing slot have been developed by birds: the alula provides a mid-wing slot, and emargination of the distal primaries provides one to several slots at the wing tip. In a highly slotted wing such as that of the Catbird (*Dumetella carolinensis*), diagrammatically shown in figure 3, the slotted length (S) may be about 55% of the total length of the wing.

With positive pressure beneath the wing and negative pressure above it, air inevitably spills over the wing tip (fig. 4), causing a loss in lift. Incidentally this circulation causes an enlarging spiral wing-tip vortex behind each wing tip, with upwelling air on the outer side. It is to use the lift from these vortices that geese and other birds fly in echelon. Use of the wing-tip vortex also unquestionably explains McIlhenny and Osborn's (1938) observation of Black Vultures (*Coragyps atratus*) sailing behind aircraft. Formation flight salvages much of the substantial amount of power lost

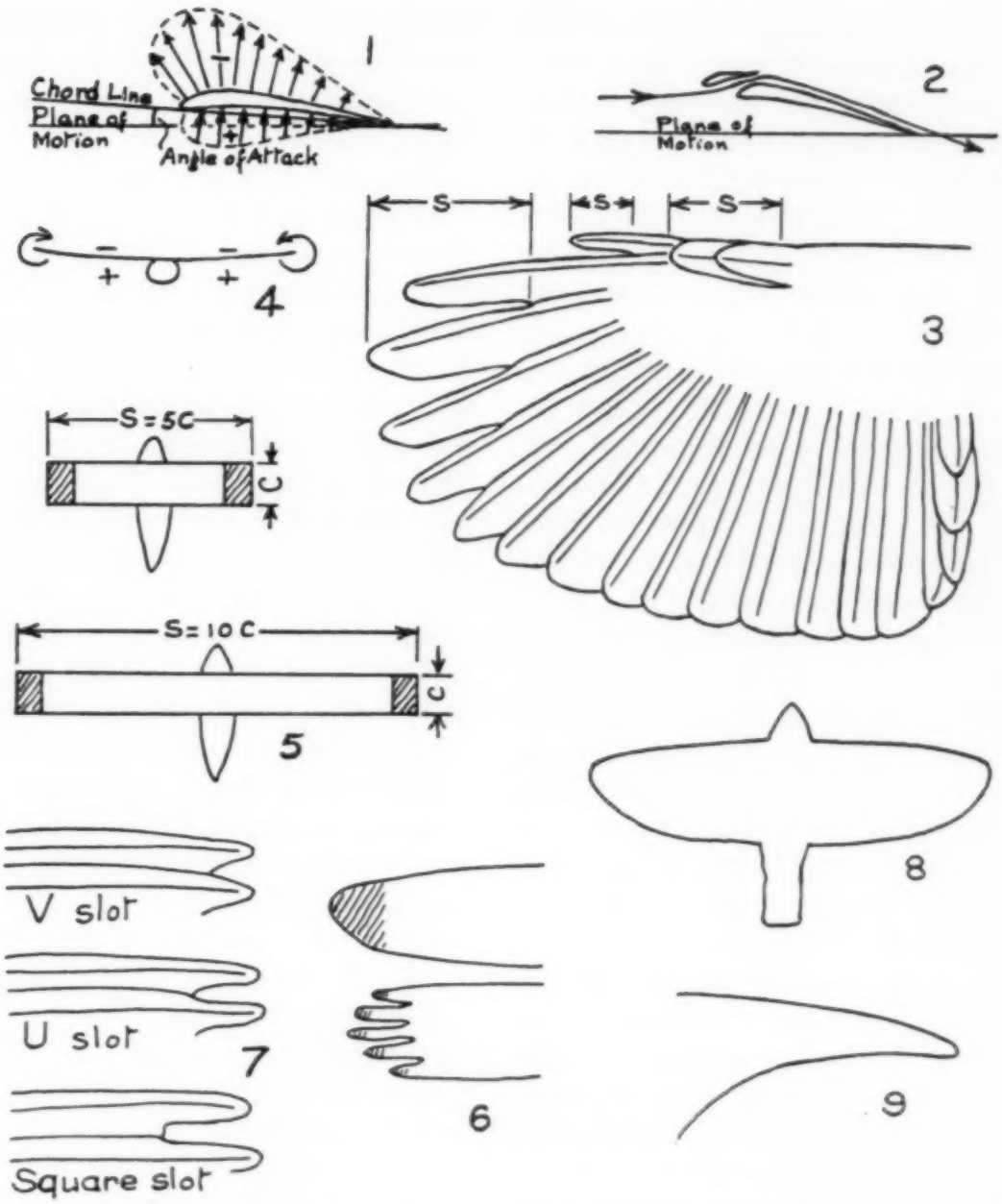

FIG. 1. Section through wing, showing approximate magnitude and direction of lift at different points on surface. Angle of attack = angle between chord line and plane of movement through air.

FIG. 2. Wing section showing action of slot in speeding flow of air over wing and delaying turbulent break-away.

FIG. 3. Diagrammatic plan view of wing of Catbird. Slotted portions (S) make up 55% of length of wing.

FIG. 4. End view of bird or airplane, showing how positive pressure below and negative pressure above wing produce wing-tip vortices.

FIG. 5. Effect of increased aspect ratio in decreasing ineffective proportion of wing. S = span; C = chord; shading shows ineffective area at wing tip. In upper aircraft, with aspect ratio (S/C) = 5, ineffective area is twice as great as in the lower one, with aspect ratio = 10.

FIG. 6. Reduction of loss of lift by slotting of wing tip. Shading represents ineffective area.

FIG. 7. Types of slot formed by emargination of primaries; primitive V slot, at top; more advanced and more efficient U slot, found in many woodland birds, center; highly efficient square slot, found in birds of prey, below.

FIG. 8. Plan view of House Sparrow, showing typical elliptical wing.

FIG. 9. Plan view of wing of American Golden Plover, a fully developed high-speed wing.

at the wing tip; but the loss can also be reduced in the individual bird in two ways: by increasing the aspect ratio or by slotting the wing tip.

By increasing the aspect ratio (ratio of span to chord) the effective proportion of the wing is increased. This relationship is shown diagrammatically in figure

5 for a rectangular wing. Suppose that the shaded area at each wing tip is functionless; it is clear that the proportional loss of effective area is twice as great in the upper craft (aspect ratio = 5) as in the lower one (aspect ratio = 10). Increase in aspect ratio is limited by requirements for structural strength and maneuverability. [In practice, wings are not rectangular and the chord is difficult to determine directly; it is therefore replaced by the mean aerodynamic chord, which is (total wing area/span); and aspect ratio thus becomes (span^2/area).] Just as the aspect ratio is basic to aircraft design, consideration of it is fundamental to an understanding of bird adaptations. In measuring wing areas, for the determination of aspect ratios and wing loadings, the wing is assumed to be continuous through the body, for the body usually supplies a little lift. There is no sound reason for departing from these standard definitions, as a few writers have done when dealing with bird flight.

The loss of lift at the wing tip can also be materially reduced by transforming the tip into a series of aerodynamic slots. Not only does slotting in itself increase lift, as we have seen, but the ineffective area becomes smaller, somewhat as shown in figure 6. The upswept emarginated primaries of a soaring eagle or vulture show us that they are delivering substantial lift. Tip slots present serious mechanical problems in large aircraft, but in birds they represent a simple and almost inevitable development; for the basic disposition of the remiges, with the distal primaries lying nearly at right angles to the longitudinal axis of the bird, was already established in *Archaeopteryx*. The development of slotting, by the emargination of some of the primaries, developed as needed in various groups of birds; and, by a suitably chosen series, we can see all stages of its evolution. It is interesting to consider that if, in the evolution of the first birds, the primaries had been established running fore and aft, at right angles to the supporting bones as the secondaries run, wing evolution would have been so restricted that birds might have been unable to invade several ecological niches.

Emargination of the primaries produces three forms of slot, which are shown in Figure 7. The V slot is the least efficient and is apparently always the first to be developed. In it the spacing is too wide for efficiency at the feather tips, and it diminishes gradually to a base so narrow that air passage through it is choked by friction. Thus only a small proportion functions effectively. The U slot is much more efficient than the V type, preserving the optimum width for most of its length, but again causing some choking at the base. This is the most advanced type of slot to be found in most groups of birds. The square slot represents a final refinement in which choking at the base is completely eliminated by a remarkable pattern of emargination. The selective advantage of the square slot is evidently negligible in most modes of life, for it seems to have developed only among the diurnal raptores and the owls, in which the carrying of prey and, sometimes, great body size have imposed special lift problems. Its clearly independent development in these two groups of birds emphasizes its efficiency.

The slots nearest the leading edge of the wing are the most important and are always best developed. In many birds we see a gradation from shallow V slots near the trailing edge to deep U or square slots at the leading edge. The rearmost, and aerodynamically least important, slot is ordinarily the most poorly developed. The graded slots in a single wing seem, therefore, to epitomize the evolution of the slot.

Graham (1930) regarded wing slots as a safety device that promotes stability in flight, but there is little to support such a suggestion. There seems to be no rational explanation for the supposition that birds in various families need a stabilizing device while those in many others do not; whereas the correlation between slotting

and the need for short wings, high lift or repeated take-offs is so close that it gives the strongest support to the view that slotting has evolved to supply added lift. Moreover, as Smith (1952) has indicated, birds started off with a high degree of static stability; but, as the development of the cerebellum permitted the detection of small degrees of imbalance, they have tended increasingly toward unstable flight with its compensating advantages of greater maneuverability and lower stalling speed. It is difficult to believe that a stabilizing device should have developed *repeatedly* in opposition to this trend.

The development of the alula is generally correlated with lift requirements in much the same way as the tip slots. It is usually well developed in large birds and in active small ones such as many passerines. It presumably developed as an aid in smoothing the contour of the wing in the region of the manus; and

TABLE 1. *Aerodynamic characteristics of representative birds grouped according to wing form*

Species and wing form	Weight (g.)	Span (mm.)	Wing loading (lb./ft.²)	Aspect ratio	Alula (mm.)	Slots (mm.)
Elliptical						
Lagopus lagopus ungavus (juv.)	468	523	1.5	4.4	55	U 78, 95, 90, 72, 67; V 36, 10
**Porzana carolina*	75	325	0.78	5.4	32	—
Corvus brachyrhyncos	361	852	0.62	6.1	63	U 70, 102, 108, 96, 57; V 6
Certhia familiaris americana	8.3	183	0.23	4.6	13	U 16, 22, 19, 13; V 6
Dumetella carolinensis	40.5	272	0.53	4.7	22	U 12, 25, 28, 19; V 12, 6
Eremophila a. alpestris	38.5	325	0.43	5.75	19	U 32, 29, 19; V 13
Dendroica petechia	8.8	175	0.28	4.9	11	U 13, 13, 13, 9; V 6
Passer domesticus	27.5	254	0.48	5.5	16	U 19, 19, 12; V 6
Molothros a. ater	48.9	310	0.58	5.7	22	U 25, 22, 19; V 13
High Speed						
**Anas rubripes*	1245	825	2.6	7.0	51	U 51; V 44, 44, 25
**Spatula clypeata*	652	735	2.0	8.1	44	U 41; V 38, 38, 13
**Falco peregrinus anatum*	1222	818	3.0	7.9	63	Sq 51; V 25
**Pluvialis d. dominica*	136	530	0.93	9.4	25	—
**Chaetura pelagica*	17.3	262	0.44	8.6	13	V 13, 6
Archilochus colubris	2.93	89	0.36	4.7	—	V 6
Petrochelidon pyrrhonota albifrons	19.1	282	0.37	7.5	14	—
High Aspect Ratio						
**Gavia i. immer*	3260	1170	5.0	10.0	25	At most V 51, 25, 19, 19
**Cygnus columbianus*	6800	1840	3.5	8.5	114	U 114, 114, 102, 25
†*Diomedea exulans*	8160	3455	2.5	?18	ca. 60	At most V 30, 20, 20
**Diomedea irrorata*	2040	2313	0.7	?15	ca. 50	Apparently none
**Larus argentatus smithsonianus*	850	1350	0.78	8.1	51	V 51, 51, 19
**Clidonias nigra surinamensis*	60	557	0.36	8.9	19	—
Slotted High Lift						
Aquila chrysaëtes canadensis	4415	1940	1.8	7.4	127	Sq 165, 203, 228, 216, 165, 51
**Buteo jamaicensis borealis*	1267	1105	1.2	5.8	83	Sq 89, 114, 127, 108; V 13
**Circus cyaneus hudsonius* ♂	414	940	0.68	7.1	76	U 70, 89, 95, 63
Otus asio naevius	156	536	0.61	5.5	38	Sq 25, 38, 38, 32, 19

* Weights are averages of published data, not those of specimens examined.
† Approximate estimates from illustrations and published descriptions.

later, as it lifted accidentally during take-off, started to function as a slot, after which its further development would have been highly adaptive in short-winged or heavy birds. The alula is conspicuously poorly developed in two groups of birds that I have had the opportunity to examine. In the loons (*Gavia*) it is extremely small and ill-formed, probably primitively so—a point to which we shall return in the final section. In the hummingbirds (Trochilidae) it is small or absent, in this case presumably secondarily so; for it is of normal proportions in the Caprimulgidae, which are probably not far from the ancestral stock of the hummingbirds. Three factors have encouraged its suppression in this family: small body weight; a high-speed wing, which cuts the air fast even in hovering; and a powered up-stroke (Savile, 1950) in which an alula is inevitably functionless.

One further elementary physical fact must be noted here. In bodies that are identical in specific gravity and shape, surface area varies as the square, but volume (and mass) as the cube of the linear dimensions. Thus, in birds that are otherwise similar, one with a 12 inch span will have twice the wing loading (weight per unit area) of one with a 6 inch span. We see at once why the upper weight limit is so much smaller for flying than for flightless birds. It is also clear that the lift problem is much more acute for large birds than for small ones.

Basic Wing Forms

Several easily recognized wing forms have been developed in the birds. Although these forms intergrade almost infinitely they form a convenient basis for consideration of the correlation of wing form and function.

Table 1 gives the principal aerodynamic data for a number of birds, grouped according to wing form. Wing loadings have been given in lb./sq. ft., which is standard aircraft practice in English-speaking countries. They may be converted to g./sq. cm. loadings by multiplying by a factor of 0.488, but the metric figures have been excluded from the table to avoid crowding.

The Elliptical Wing

The elliptical wing of aircraft terminology is not truly elliptical, for it is not symmetrical about its long axis. However, it approaches an ellipse, there is no word that precisely expresses its form, and the term is unambiguous in aerodynamic context. Although its proportions vary substantially its general appearance is distinctive. Figure 8, which represents the plan view of a House Sparrow (*Passer domesticus*) in the altitude of level flight, provides a good example of the elliptical wing. This outline is almost exactly that of the wing of the Mark II Spitfire of immortal memory; but of course the Spitfire, adapted to much higher speeds, had a very different wing section and a pronounced wing root fairing. Construction cost has limited use of the elliptical wing in aircraft, but there has been nothing to hinder its evolution in birds.

At low and moderate speeds (i.e., below the point where air compressibility becomes an appreciable factor, at about 350–400 m.p.h.) this wing plan is very efficient. Wing-tip turbulence is slight, and pressure distribution is unusually uniform over the wing surface regardless of altitude. Lift is accordingly high and control dependable under a wide variety of circumstances. Thus this shape, together with low aspect ratio, gives good maneuverability. It is not surprising that this form of wing, with various elaborations, has been widely adopted in the Passeriformes, most of whose members must move easily through restricted openings in vegetation. Essentially the same wing form is found in other birds of similar habitats, e.g., Galliformes, Piciformes, Columbidae and the woodcocks (*Philohela* and *Scolopax*). This form is aerodynamically advanced and is undeniably well adapted to birds of forest and shrub

habitats, but whether it must be considered advanced in the evolutionary sense is less certain. We shall return to this point later.

Slotting is developed to varying degrees in birds with elliptical wings. In the larger birds, such as most of the Galliformes and Corvidae, the slots are well-developed and are kept open in level flight: hence the "ragged fingers" of the crow, more admired by the aerodynamicist than the poet. In many but not all of the smaller passerines slots are fewer and in part V-shaped. Their development is correlated with activity as well as size. Thus we find that slots are generally better developed in the wood warblers (Parulidae) than in more sedentary passerines of more than twice their weight. Compare, for example, the Yellow Warbler (*Dendroica petechia*) with the House Sparrow (*Passer domesticus*) and the Cowbird (*Molothros ater*) in table 1. The Brown Creeper (*Certhia familiaris*), which, like the warblers, must make thousands of take-offs each day in feeding, is also very highly slotted for its size. Take-off consumes much more power than level flight; and abundant slotting, to give high lift at low speed, has been developed to allow repeated take-offs with moderate power consumption. No figures are available, but a warbler probably consumes as much power in an hour of active feeding as in an hour of steady flight on migration.

An extreme case of slotting is seen in the Catbird (*Dumetella carolinensis*) (fig. 3 and table 1), which inhabits unusually dense shrubbery for a bird of its size. Its habitat demands short wings. Adequate lift with a very small span is secured in the Catbird by the development of a wing with a deep chord, pronounced camber, a substantial alula and a well graduated series of tip slots. It will be seen from figure 3 that the short distal primary serves to slot the area of wing between that served by the tip slots and that served by the alula.

The figures for wing loadings do not fully indicate lifting power, which is partly dependent on slotting. These figures, and others not reproduced here, do show, however, that, although wing loading generally falls with decrease in body weight, it does not fall as fast as one might expect. If the American Crow, weighing about ten times as much as the Yellow Warbler, were simpler a large-scale model of it, it would have a wing loading ten times as great instead of a little over twice as great. Thus the smaller birds are adapted for flying in confined spaces by being able to possess disproportionately small wings without having unduly high wing loadings. In the evolution of the woodland passerines the advantages accruing to small body size seems to have exercised considerable influence.

If the high wing loading and lack of slotting shown for the Sora (*Porzana americana*) are characteristic of the rails, we see why they take off so laboriously. Once airborne the rails are capable of long migratory flights and even trans-oceanic passages, but their feeding habits have not encouraged the evolution of devices to ease the labor of take-off.

Although the tip slots of large birds are generally open in level flight, this does not seem to be generally true in small birds, in which they are usually closed to reduce wing area and increase speed. At take-off and landing the tail is spread and lowered as a landing flap, and, to compensate for the upward tail moment and keep the center of pressure over the center of gravity, the wing is drawn forward by a pull on the tendon running out to the first digit. This action slightly deepens the chord of the inner part of the wing; moves the first digit to open the alula slot; pulls the manus forward, stretching the elastic ligament to separate the distal primaries and open the wing-tip slots; and, often at least, so stresses the wing bones that they arch slightly, increasing both camber and lateral camber (decrease of dihedral from root to tip). All these associated changes help to in-

crease lift, but the opening of the slots is particularly effective.

The High-speed Wing

The high-speed wing is characterized by a much lower camber (flatter section) than is general in bird wings, moderately high aspect ratio, taper to a relatively slender elliptical tip, pronounced sweepback of the leading edge and sometimes even of the trailing edge, lack of tip slots (except in the falcons, which must be able to carry loads, but in which the slots are closed in rapid flight), and development of a pronounced fairing at the wing root to blend the trailing edge of the wing into the body. Figure 9 shows the plan view of the wing of the American Golden Plover (*Pluvialis dominica*), a typical example of an advanced high-speed wing. This wing form is highly developed in the falcons, plovers and sandpipers, swifts and hummingbirds, and swallows. It is less highly developed in the ducks; and it is approached in the terns and jaegers which also tend to the next category. In these six groups the development of the high-speed form must have been essentially independent. Even if all these birds arose from ancestral forms of open habitats, in which a primitively long wing was permissible, such characters as sweepback and the wing root fairing must have developed separately.

The similar external form is, of course, often accompanied by profound anatomical differences. Thus the swifts (ecologically similar to the swallows) and the hummingbirds have a short and massive humerus, radius and ulna with heavy supporting tendons, in contrast to the slender bone structure of the swallows; the difference being correlated with an entirely different wing action (Savile, 1950). Most of the characteristics of the high-speed wing are easily explained. Low camber and a short chord reduce drag; a moderately high aspect ratio conserves power for prolonged flights; the elliptical tip contributes to pressure equalization; the root fairing reduces turbulence, and consequently drag, at the junction of wing and body. Only the pronounced sweepback is seemingly unexplained by orthodox aircraft aerodynamics; at least enquiry of several aeronautical engineers and statement of the problem in print have brought no explanation. I suspect that the answer may lie in some peculiarity of airflow over small wings that does not show up in airplane prototypes or large-scale models. Its repeated occurrence in, and only in, fast fliers emphasizes its evident importance. Its function is not to be confused with that of sweepback in modern aircraft that operate in the air-compression speed range of about 400 to 700 m.p.h., in which it serves to postpone the shock stall. Not even the fastest swifts or falcons exceed 200 m.p.h.

Table 1 shows that wing loadings in birds of the high speed group are generally higher than in birds of similar weights in other groups. This tendency is to be expected for at high speed a wing provides more lift per unit area than at low speed, and unnecessary area simply increases drag. The wing loading for the Ruby-throated Hummingbird (*Archilochus colubris*) seems very high for so small a bird, but it is probably representative of the family. A hummingbird's mode of feeding demands that it hover with the minimum oscillation of the body. This end is achieved by having very small wings beating very fast. The inertia of the bird's body is thus sufficient to keep its movement much below that commonly seen in other hovering birds.

Among the ducks I find that the diving ducks (Aythyinae) consistently have higher wing loadings and less slotting than dabbling ducks (Anatinae) of comparable size. These differences are correlated with habit. The dabblers are often seen in small ponds, sometimes tree-fringed, and all can spring directly out of the water into a steep climb. The divers generally occur in more open water and, except for the little Bufflehead (*Bucephala albeola*), all North American spe-

cies must make a pattering take-off and a shallow climb.

The High-aspect-ratio Wing

The high-aspect-ratio wing in its full development is confined to oceanic soaring birds, on which, unfortunately, I have been unable to assemble much quantitative data. We have seen that lift may be increased by raising the aspect ratio. In aircraft the ultimate development of this form is seen in sail-planes, in which a figure of 12 to 18 may be found, the former where some maneuverability is needed, the latter for extreme endurance. Aspect ratios of 10 or slightly more may also be found in airplanes designed for great range and fuel economy; but structural limitations prevent much higher figures. A very high aspect ratio is obviously impractical for any land bird in landing and taking off; for woodland birds it is completely impossible. But in sea birds that take off only from the sea or from a cliff, from which they may launch themselves without preparatory wing beats, a very long wing is the best means of securing high lift. This wing form is most fully developed in the albatrosses, frigate-birds and tropic-birds, but is also well marked in the gannets and gulls. In none of these birds do we find any appreciable development of tip slots, although the alula may be of substantial size. The lack of well-defined slots might be due to genetic deficiency but it is probably attributable at least in large part to greater aerodynamic efficiency of a high aspect ratio. The latter explanation is supported by the occurrence of high aspect ratio repeatedly in birds whose habitats permit it.

Soaring is seldom practiced by small birds, except in unusually strong updrafts generally of mechanical origin, despite the fact that their small size gives them a low wing loading. The principal explanation seems to be that at the low rates of airflow commonly found in soaring (much lower than across a beating wing) the high proportion of edge seriously reduces the efficiency of the small wing. We must also remember, however, that soaring is less likely to be developed by the smaller birds because powered flight is less strenuous for them than for the larger species.

The data given in table 1 for the Wandering Albatross (*Diomedea exulans*) and the Galapagos Albatross (*D. irrorata*) have been compounded from various available weights, measurements and illustrations, largely from Murphy (1936). Dr. A. L. Rand has kindly supplied alula lengths and wing tip diagrams from skins in the Chicago Natural History Museum. The smaller albatrosses seem to be without functional tip slots, but there is a suggestion of emargination in the outer primaries of the Wandering Albatross. Clearly there must be an upper size limit for soaring birds, because wing area cannot be indefinitely extended to keep pace with weight, which, we recall, increases as the cube of the linear dimension. The figures given are for a large specimen of the Wandering Albatross. The high wing loading suggests that this species has just about reached the practical limit of size; for it is doubtful if the aspect ratio could be raised much further without the wings becoming unmanageable. It is probably not due to chance that this species and the almost equally large Royal Albatross (*D. epomophora*) are practically confined to latitudes 30°–60°S, the region of strong and almost continuous southern westerlies in which mechanical up-drafts are generally powerful. In contrast, several of the smaller albatrosses range north through the doldrums and horse latitudes where air circulation is generally weak. Parenthetically it may be noted that even when a calm is experienced in the southern westerlies region a tremendous ocean swell persists with rollers perhaps à mile or more from crest to crest and of great height. Those who have put off in a ship's boat under such circumstances tell of the ship being completely hidden from their view when the boat is in a trough.

These great rollers make mechanical updrafts that must be adequate to support a Wandering Albatross once it is aloft, and supply a more plausible explanation of its soaring under calm conditions than some of the curious schemes that have been propounded. The real difficulty is for a large albatross to take off in such a calm, and the apparent rudimentary slots of the Wandering Albatross may be of some value in these circumstances. Bergmann's rule, that in warm-blooded vertebrates related forms tend to be larger in cold than in warm regions, is generally thought to be based on the greater thermodynamic efficiency of large body size; but this rule can only be applied safely below the specific level (see, e.g., Mayr, 1956), and I feel certain that the distribution of the albatrosses has primarily an aerodynamic rather than a thermodynamic basis.

Of the species examined, the gulls have aspect ratios of about 8 to 9, and the terns 9 to 11. The terns do not have a typical soaring wing but show a tendency toward the high-speed type, notably in the possession of a small wing-root fairing. This intermediate form is reflected in their habits, for they use powered flight more than the gulls and some species make long migration flights. The gulls and terns are much less pelagic than the typical oceanic soaring birds and their moderate aspect ratios are a compromise between aerodynamic efficiency and the needs of a partly terrestrial habitat. The jaegers (*Stercorarius*) also approach the high-speed wing form.

Two other birds have been included in the high-aspect-ratio section of table 1 simply because they fit in no other category. The Whistling Swan (*Cygnus columbianus*) has a body weight equalled or exceeded by relatively few birds that fly strongly. It has a high aspect ratio, which, together with formation flying, conserves much power on long migration flights. It also has a large alula and substantial tip slots, which must greatly assist the bird in taking off and, by reducing stalling speed, ease the shock of landing. A similar compromise between high aspect ratio and slotting is seen in other birds whose habitats justify it. The geese have wings somewhat similar to those of the swans, much of the length being provided by the primaries. The pelicans have most of the wing length provided by the humerus, radius and ulna, but the primaries provide an array of well-formed slots that remain open in level flight. These two groups provide a good example of the same aerodynamic end being provided by quite distinct anatomical means.

The Common Loon (*Gavia immer*) is a flying anachronism that fits into no aerodynamic category. It will be discussed in the final section.

The Slotted Soaring or High-lift Wing

Among birds that inhabit forested areas or other impeded terrain a soaring wing of high aspect ratio is inadaptive. A slotted soaring wing of moderate aspect ratio has been adopted as an alternative. It is conspicuously developed in the vultures, eagles and buteos, ospreys, and harriers; but the same tendency is seen in other, unrelated large birds such as the Raven (*Corvus corax*) and the pelicans, discussed in the preceding section. It is also reasonable to include the owls in this category; for, although they seldom soar, their wings have evolved similarly to develop exceptionally high lift. The eagles, ospreys and harriers, of relatively open habitats, seem usually to have appreciably higher aspect ratios than the woodland hawks and owls, but I have not been able to examine enough species to be sure that a correlation with habitat is invariable.

The low wing loadings of these generally large birds show us how large their wings are, but it is in their slotting, alulas, and pronounced camber and lateral camber that their great efficiency at low speeds is revealed. It should also be noted that the degree of slotting is markedly greater in proportion to size in the large than in the small birds of this group,

a relationship that is conspicuous in all other hawks and owls examined as well as those tabulated. It seems to be quite clear that in both these groups of predators slotting has continued to develop as long as its increase remained appreciably adaptive. I know of no indication of any species of raptores being deficient in adaptive mutations bearing upon wing form.

The Problem of the Primitive Wing

Consideration of these various adaptations in the wings of contemporary birds leads naturally to speculation about the early evolution of the wing and as to what types of wing might properly be called primitive. Perhaps no existing birds can be said to have a primitive wing in the strictest sense of the word; but the loons immediately spring to mind as possessors of an outstandingly inefficient wing, and they merit some consideration. The Common Loon has an appallingly high wing loading, a very inefficient wing form with a slender and sharply pointed tip, small slots of very poor form, if indeed the vague emarginations of the primaries function as slots, and a minute alula just the length of that of a 130 g. Bronzed Grackle (*Quiscalus versicolor*) but much less efficiently shaped and probably only functioning to smooth the wing contour. Under these handicaps the Common Loon takes off only with difficulty, after a long run, and climbs shallowly. It is thus debarred from small and tree-fringed or steep-sided ponds that might otherwise provide good nesting sites. The smaller Red-throated Loon (*G. stellata*) and Pacific Loon (*G. arctica pacifica*) evidently have substantially lower wing loadings, for they take off with less labor, but they also are somewhat restricted in their breeding sites. The Yellow-billed Loon (*G. adamsi*) I have not seen in the field, but I suspect that its performance is comparable to that of the Common Loon. Once airborne a loon can only fly fast, and its landing stresses must be tremendous, which may partly account for its heavy body feathering.

It has been suggested to me that the loon's wing form is hydrodynamically advantageous, but I know of no supporting evidence. The loons feed predominantly on live fish and must swim fast under water. They swim by alternate strokes of the feet; and, since *Hesperornis* had the same elongated patella as *Gavia*, it seems probable that the whole lineage has swum in this manner. The concensus of opinion seems to be that the loons do not use their wings beneath the surface. They are so heavy-bodied that compression of the body plumage must give them approximately neutral buoyancy. Thus even when they do feed on the bottom the need for muscular activity to keep them down must be small and is probably applied through the feet. If the wings are opened at all under water they probably trail slightly and serve as hydrovanes to control the level of the swimming bird. Even if the wings were actively used in bottom feeding, somewhat as in the diving ducks, the pointed tip would represent a significant loss of power. At take-off the long, pointed tip will drag more in the water than a shorter, broader and rounded tip such as that of a duck. At best, therefore, the loon's wing form is hydrodynamically neutral if the wing is kept tightly closed, and is disadvantageous if it is opened at all under water.

Although it is possible that the loons have at times lived under conditions in which efficient flight was of minor importance (perhaps, like their Cretaceous relative *Hesperornis*, inhabiting warm inland seas), today their inefficient flight is an undeniable disadvantage. Not only do they make long migration flights; but, in the arctic, islets suitable for nest sites are often to be found only in ponds too small or too shallow to provide an adequate supply of fish, and daily feeding flights are necessary. If the loons were on the verge of becoming flightless, one might expect that the wing would become shortened and rounded, rather than nar-

rowed, thus interfering less with the birds' swimming. Such shortening with a rounded tip seems to be general among flightless birds. Moreover an effective alula, had the loons ever possessed one, could not interfere at all with swimming and would be advantageous as long as flight continued. It thus seems to be a tenable hypothesis that the loons have a primitively inefficient rather than a degenerate wing, for its form seems to be inadaptive both for flying and for swimming.

Despite its teeth and lack of wings, *Hesperornis* so closely resembles the modern loons in many skeletal characters that it is probable that loon-like birds, capable of flight but already well adapted to an aquatic existence, must have existed appreciably earlier. The wing degeneration of *Hesperornis* might have occurred abruptly by a major mutation, but the great body size and the slenderness of the humerus (all that was left of the wing) suggest a long evolutionary period after flightlessness set in. However, as Dr. G. G. Simpson (*in litt.*) points out, paleontology shows that such specialized forms have sometimes developed quite rapidly, and the meager fossil record of Mesozoic birds leaves the chronology of the loon ancestors in doubt. It seems clear, at least, that flying birds ancestral to both *Gavia* and *Hesperornis,* and look-like in general form, must have existed in early Cretaceous times, possibly not very much later than the upper Jurassic *Archaeopteryx.*

Archaeopteryx (considered here as including *Archaeornis,* the Berlin specimen in which the wings are well preserved) had a wing entirely unlike that of a loon. The eight primaries are graduated proximally and distally to form a nearly elliptical wing tip of surprisingly modern appearance (Savile, 1957). The wings were not preserved in fully extended position, but they seem to have been nearly elliptical in outline, and, except for the lack of an alula or emarginated primaries, are not unlike those of many passerines in general appearance. De Beer (1954) lists the lack of a keel on the sternum, the structure of the crista pectoralis and the short coracoid as evidence that *Archaeopteryx* was at best a weak flier; and to these indications we may add the unstiffened elbow joint, quite incapable of withstanding a powerful down stroke. Nevertheless the wing is appreciably advanced aerodynamically and *Archaeopteryx* was probably quite a successful glider. Even if we accept Heilmann's (1926) suggestion that the graduated primaries evolved as an aid to the bird in clambering about in trees, which is plausible but far from proven, it seems beyond question that aerodynamic refinement had operated for a substantial time to produce the *Archaeopteryx* wing.

It is unlikely that the loons are close to being lineal descendants of *Archaeopteryx,* for the skull of *Hesperornis* is in some respects less advanced, e.g., the splenial is conspicuously developed on the outside and the mandible bears many more teeth than that of *Achaeopteryx.* Moreover the upper teeth are all in the maxillary rather than being divided among the maxillary and premaxillary as in *Archaeopteryx.* If Heilmann's suggestion that the distal tarsals were fused with their respective metatarsals but not with each other is correct, *Archaeopteryx* does not seem to be directly ancestral to any modern birds. It may have split off from the main avian stem before any particular wing form became firmly established. I visualize it as an evolutionary offshoot from the presumptive pro-avis in which the digits of the fore-limb remained unreduced because the species was genetically incapable of developing the modifications needed for successful flight. Successful fliers may have existed contemporaneously with *Archaeopteryx,* but they would have been much less likely to fall victim to the conditions that caused the preservation of the two *Archaeopteryx* specimens.

It is accordingly suggested that the pro-avis gave rise to the *Archaeopteryx*

line on the one hand and to the main avian stem on the other; that in the main avian stem there was a rapid reduction of the manus as a rigid anchorage for the primaries to withstand the stresses imposed by the developing flight muscles; and that very soon afterwards, probably in late Jurassic and before any particular wing form became firmly fixed, the main stem split into two branches, an aquatic one leading to the loons (and perhaps some other aquatic groups such as Steganopodes and Tubinares) and a terrestrial-arboreal one leading to the landbirds in general and to some that are secondarily aquatic. In the aquatic branch there would have been nothing markedly deleterious in the production, through elongation of the distal primaries, of a long, slender wing, which seems to be the simplest and fastest solution to the problem of increasing lift.

This scheme is, of course, an oversimplification. There need not have been a single sharp division, but perhaps several divergences in each direction from a group of unspecialized ancestral species. There has probably always been, and still is, a tendency among genetically plastic birds for the wing to lengthen when the habitat permits it and to be shortened and develop other high-lift devices when the habitat makes a long wing deleterious. That many groups of birds are genetically capable of shortening their primaries is shown by the wing reduction that has occurred in various flightless birds and by recorded mutations in the domestic fowl. Evidence for the reverse tendency, when elliptical-winged birds revert to open habitats, is less abundant, but the occurrence of high aspect ratios in several unrelated groups of open-ground or oceanic birds strongly suggests that the tendency is widespread. The Horned Lark (*Eremophila alpestris*) is perhaps in process of elongating its wing after reversion to an open habitat. The longest primaries in this species project beyond the elliptical curve of the rest of the wing in a most curious manner that is difficult to explain on any other basis. Presumably the inner primaries would eventually increase in graded proportion, to restore the smooth curve of the wing, if no further reversion to a less open habitat occurred. The inference from this example of the lengthening of a well-formed elliptical wing is that high aspect ratio is aerodynamically preferable to wing slots with a short elliptical wing, and will generally be adopted when mechanical considerations permit.

According to the explanation presented above many structures in the ancestral loons at first underwent tachytelic evolution, which soon gave way to bradytely. For the wing, however, it is suggested that the loons early ran out of such adaptive mutations as continued to occur widely in some of the presumptively related sea birds. At first the wing loading of the loons may not have been excessive: it may actually have increased as time went on, because specialization for swimming under water tends to increase specific gravity of the body, since excessive buoyancy causes power to be used to keep the bird down and reduces speed. As body weight increased, the genetically rigid wing failed to develop in compensation.

In the terrestrial-arboreal branch interference from long primaries would have been intolerable and would quickly have forced the evolution of shorter and deeper wings. Aerodynamic advantages would quickly transform such a wing into elliptical form; but aerodynamically directed similarity between the wing of *Archaeopteryx* and those of any early flying birds does not indicate genetic relationship, any more than their functionally similar wings imply a genetic relationship between a grouse and a Catbird.

It has perhaps been generally accepted, following Heilmann's arguments, that the pro-avis was fully arboreal, principally because the arboreal habit best explains the long fore-limbs and opposable hallux; but I see no reason to assume that the pro-avis was as completely arboreal as *Archaeopteryx*. The pterosaurs, with

their long wings and unspecialized feet, probably developed as gliders launching from steep slopes or cliffs. It is quite possible that the pro-avis was intermediate in habit, with a partly opposable rather than a fully opposed hallux. The concept of such a pro-avis of moderately open habitat in which the first solution of the lift problem was the easiest one, i.e. elongation of the outer primaries—*a solution favored by all contemporary birds of open habitats*—makes the early evolution of loon-like birds and the form of the loon wing much easier to visualize than do Heilmann's strictly arboreal pro-avis and the postulate that *Archaeopteryx* was almost directly on the main avian stem. As a less specialized organism such a pro-avis also strikes me as the more plausible ancestor for a large and diversified group of organisms.

The scheme that I have outlined is, of course, highly speculative, but in most respects it is no more so than others aimed at explaining early evolution of the birds. The wing of the loons is so astonishingly inefficient in form that I cannot believe it can have evolved (of necessity by a whole series of deleterious mutations) from a wing of more normal form. The proposed scheme is the only way in which I can account for the loon wing on the presumption that it is primitively inefficient rather than degenerate.

Summary

Some of the aerodynamic aspects of the wings of birds have been discussed and examples of adaptive evolution given. For convenience of discussion wings have been grouped into four categories: the elliptical wing, adapted to operation in confined spaces; the high-speed wing, developed independently in several groups of birds that feed on the wing or make long migration flights; the high-aspect-ratio wing, found generally among oceanic soaring birds; and the slotted high-lift wing, found in terrestrial soaring birds and predators that must carry heavy loads. Each wing form has been developed by several unrelated groups of birds, emphasizing the fact that wing form is usually genetically plastic. The loons possess a wing that, alone of all birds studied, does not fall into any established category or into a position intermediate between any two, and which is so poorly formed aerodynamically that its form is believed to be primitive. An evolutionary scheme that embraces this postulate has been presented.

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THE RELATION OF AN INVERSION SYSTEM TO RECOMBINATION IN WILD POPULATIONS[1]

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INTRODUCTION

A chromosome inversion results in an alteration of the original gene order. The alteration produced by each inversion is readily recognizable in the salivary gland chromosomes of *Drosophila* because of the constant differences in appearance and arrangement of the bands that characterize them. This genus is accordingly a facile subject for a study of the geographical distributions and frequencies of different arrangements in wild populations. The number, kinds and frequencies of arrangements vary from species to species (Patterson and Stone, 1952; da Cunha, 1955). The inversions of *Drosophila pseudoobscura*, which we propose to discuss, are confined chiefly to two chromosomes, the acrocentric third and the metacentric X and, in the latter, to its right arm. Nineteen arrangements have been recorded in the third chromosome and another is inferred to have existed at one time, if it does not now. They generally overlap each other and are only rarely included, and their heterozygotes are correspondingly complex. Twelve arrangements of this chromosome are known in the populations we shall discuss. Only three inversions have been recorded in the right arm of the X. They are structurally independent of each other, but are not apparently separable by crossing over in nature, and thus form only one effective arrangement. It is associated as a unit to some genetic factor that causes an alteration of the sex ratio and produces mostly females in matings of males that carry it. Three inversions have been found in the second chromosome and one in the fourth. They occur only in low frequencies in the area we shall discuss, if at all, and can have but little effect on the operation of the inversion system. Five types of the Y chromosome are associated with these inversions but their relation to the system is unknown. They may need be taken into account ultimately. As far as known, only one type occurs in this area. Thus, the inversion system of *D. pseudoobscura* is more or less equally divided between two parts of the genome. The basic data for the whole system have been presented by Dobzhansky (Dobzhansky and Epling, 1944). More recently discovered arrangements are discussed by Epling and Lower (1957).

Dobzhansky, Holz and Spassky (1942) have estimated the relative lengths of the five chromosome arms. The third chromosome constitutes approximately 18 per cent of the total and the right arm of the X approximately 23.5 per cent. Thus, when arrangements are heterozygous simultaneously in both chromosomes, crossing over will be greatly reduced in about 40 per cent of the euchromatin of the cell concerned.

The arrangements of the third chromosome have been given names suggested by the places where they were first detected, such as Arrowhead (AR), Chiricahua (CH), Treeline (TL), Pikes Peak (PP) and Standard (ST). Those of the X chromosome are known as Standard (STX) and sex ratio (SR). The term Standard in both instances means only that these arrangements were taken as a base for comparisons.

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The frequencies of the arrangements of both chromosomes differ regularly over wide areas (Dobzhansky and Epling, 1944). Some arrangements of the third chromosome are found chiefly in the western United States. Some are found only in Mexico and Guatemala, but the predominant ones there are different from those that predominate in the United States. In the United States, where data are most abundant, the relative frequencies of the chief arrangements appear to be correlated with major climatic regimes. Thus, ST, AR and PP range from California to Texas but differ in frequency along this transect (figures on pp. 123, 124 in Dobzhansky and Epling, 1944). ST is predominant along the Pacific Mountain System from British Columbia to California, accompanied chiefly by AR and CH. Its area of predominance includes the coastal plains and valleys and the cordilleras of the Cascade and Sierra Nevada ranges of which the biota is governed by a mediterreanean type of climate of relatively mild temperatures, of winter rains and summer drought, which coinsides with the highest temperatures. The frequency of ST diminishes rapidly eastward. AR is predominant in the Intermontane Plateau, accompanied by ST, CH and PP in low frequencies. This basin is shielded by the Pacific Mountain System from winter storms from the west and by the Rocky Mountain System to the east. It is an arid region with a continental climate that reaches its extremes in the Sonoran desert. It is influenced by the attenuated winter cyclones from the west and by summer showers from east and south, coupled with high summer temperatures and cold winters. PP is predominant in Texas and adjacent Mexico where the influence of the Gulf climate is greatest and where rainfall coincides largely with the warmest months, resulting in high humidity during summer and relative drought during winter. The geographical and ecological clines of the arrangements of the third chromosome suggest different adaptive qualities of each.

The second component of the inversion system of *D. pseudoobscura,* the SR arrangement of the X chromosome, reaches its highest frequency in Arizona or the North Mexican plateau and diminishes northward and probably also southward from this center (Dobzhansky in Dobzhansky and Epling, 1944; Wallace, 1948). This region probably has the greatest extremes and variability of weather of any in the species area. It is also probably the most recently occupied because of the late Cenozoic development of its present biota (Epling in Dobzhansky and Epling, 1944; Axelrod, 1950). In California the frequency of SR is highest in southern California. The clines of frequency of the arrangements of the third and X chromosomes do not therefore coincide. The former has a longitudinal pattern in the United States, the latter a latitudinal or perhaps a radial one centered in Arizona or the North Mexican Plateau. The result in the United States is a regularly changing system of frequencies of SR with each of the predominant arrangements of the third chromosome.

These major geographical regularities associated with broad climatic regimes are reflected on a smaller scale by ecoaltitudinal clines (Dobzhansky, 1948). That in the San Jacinto Mountains is illustrated in Table 1 in which the mean gametic frequencies of the six principal arrangements during two periods are listed. The data for the first period are taken from Dobzhansky (1943). ST decreases regularly with altitude, being replaced chiefly by AR and CH. PP and TL may also decrease, but their frequencies are too low to be certain of this. SR on the other hand, does not apparently change regularly along this cline. Comparison of the means of earlier and later periods of Table 1 shows that the cline of frequencies in the third chromosome has had a considerable permanence. It is associated with an environmental gradient that will be described later.

These regularly changing frequencies

TABLE 1. *Comparison of the mean gametic frequencies of the arrangements indicated between the periods 1939–1942 and 1952–1956*

	ST		AR		CH		PP	TL	SR
	1939–1942	1952–1956	1939–1942	1952–1956	1939–1942	1952–1956	1952–1956	1952–1956	1952–1956
Pinon Flat	.416	.477	.249	.215	.280	.154	.087	.053	.116
Vandeventer Flat	—	.451	—	.230	—	.175	.088	.055	.153
Keen Camp	.325	.360	.234	.256	.394	.282	.066	.036	.113

in space are being continually modified from year to year and by long term fluctuations, or "directional" changes (Dobzhansky, 1947) of considerable regularity that occur over wide areas. The long term fluctuations that have taken place in the San Jacinto Mountains since 1939 will be discussed in another publication (Epling and Lower, 1957). For the present it is sufficient to say that each arrangement in the third chromosome fluctuates more or less independently of the others and that their fluctuations are not correlated very closely with the fluctuations of SR.

We can therefore conceive of an inversion system in *D. pseudoobscura* in which two elements form an ever changing system of relative frequencies from place to place. Each population has a distinctive system of frequencies that is associated with a different environment. Each has a considerable permanence, even though each may fluctuate from year to year as the local environment changes. This association of frequencies and environment is strong presumptive evidence that the inversion system is a means of adapting each population in some way to different and changing environments.

Regular seasonal rhythms of relative frequency are also met with in some populations. This phenomenon was first noted by Dobzhansky (1943) at Pinon Flat and Andreas Camp in the area being studied. It is the principal object of our discussion because the data we shall present suggest a way in which the adaptive function of the inversion system may operate as a mediating agent of gene recombination and the adaptability of the population.

METHODS OF COLLECTION AND ANALYSIS

Two methods of collecting have been employed. Prior to the summer of 1953 baited paper or plastic cups were set out along a fixed trapline at each station, collections being made at short intervals during the collecting period each day by using a funnel and vial over the cup (Dobzhansky and Epling, 1944). Since that time incompletely covered 2 gallon cans, with bait, have been left out for one or more days, depending on the season, after or during which the flies were collected by funnel and vial placed over a hole in the lid. The areas sampled by the second method may have been somewhat smaller than those sampled by the trap-lines. The differences in method have not seemingly altered the results. Whichever the method, an attempt has been made to procure monthly samples throughout the year. Many samples made in winter or during periods of heat and drought have been too small to be statistically useful. Nevertheless, we believe that the series of samples of the following tables is fairly representative.

The frequencies of the arrangements of the third chromosome have been ascertained in two ways, by examining the salivary glands of a single larva from the eggs laid in the laboratory by each wild female; or by examining up to eight larvae from a cross of each wild male to a laboratory stock homozygous for ST. The laboratory stock used in the second method has carried the markers *or pr* in

order to make sure that STST homozygotes are not increased experimentally as would be the case if the females were not virgin. If they are not virgin, the fact can be detected by the white eyes of the progeny. Only the results from the use of males are being reported. This method permits determination of the structural combinations such as STST, ARCH, present in each wild adult male. We have been unable to ascertain their frequencies in adult females because we have found no satisfactory way of desemination. The possibility exists that systematic differences of frequency may occur between the sexes, as has been indicated for *D. robusta* by Levitan's observations (1951). If so, the patterns of frequency we shall discuss would not be representative of the whole population. During 1952, however, we analyzed and compared gametic frequencies of the third chromosome of both males and females. Comparison showed differences in each sample, caused in part by sampling error and the differences in method, but the trends of frequency were similar. In 1956 we made a similar comparison of the SR arrangement in males and females, which will be reported on in another connection. The trends were likewise similar. We assume, therefore, that the frequencies to be reported are valid for the whole population from which the males were obtained. In determining the frequencies in the males, structural homozygosity in the third chromosome was inferred if the eight larvae examined carried the same pair of arrangements, one of which, of course, would be ST. Thus, the probability of mistaking a heterozygote for a homozygote would be 1/127. Determination of the frequencies of the SR arrangement has been direct. The laboratory females to which the wild males were mated were homozygous for the ST arrangement of both the third and X chromosomes. When the third chromosome arrangements had been ascertained by inspection of the salivary gland chromosomes, the progeny of the cross was allowed to pupate and hatch. If the proportions of males and females were more or less equal, it was assumed that the wild male had carried ST-X. If no males were present, or almost none, then it was assumed that it had carried the SR arrangement.

The Seasonal Rhythm

The raw data for each sample of wild males are presented in tables 2–4. The collections have been grouped by month in order to obtain a fairly regular series of monthly mean values throughout the year. The gametic frequencies of the six principal arrangements have been computed from these data and are given in the columns on the right. The frequencies of seven other arrangements of the third chromosome, which form less than one percent of most samples, have been omitted from the raw data. They will be presented in another paper (Epling and Lower, 1957). One of these rare arrangements, Olympic, was used in the computations for 1953 and 1954, which will account for small discrepancies between the computed values and the raw data for these years.

The pertinent data, to illustrate the seasonal rhythm of frequencies in wild males, have been abstracted from tables 2–4 and graphed in figures 1–3 for Pinon Flat, Vandeventer Flat and Keen Camp, respectively. The mean rhythm for the whole period of observation at Pinon Flat is graphed in figure 6 against the background of the seasonal rhythm of weather. Referring now to the middle range of figures 1–3, one will find the monthly frequencies of ST for each year since 1952 shown by the lighter lines, and the means for the whole period shown by the heavier. At Pinon Flat ST decreases from February or March to June and then increases to its former level during July and August. The level of frequencies attained by this time then continues, on the average, until the following March. The structural homozygote of this arrangement, STST, follows

TABLE 2. *Samples taken at Pinon Flat, 1952–1956*

Collection date	No. males	Numbers of zygotic combinations									Gametic frequencies					
		STST	STAR	STCH	ARAR	ARCH	CHCH	ST het.	All het.	SR	ST	AR	CH	PP	TL	SR
Jan. 15, 1954	14	3	1	3	—	—	—	9	11	3	.536	.071	.143	.214	.036	.214
Feb. 1–10, '54	82	31	15	7	3	—	—	38	48	6	.610	.177	.049	.091	.061	.073
2–3, '53	222	80	59	20	2	5	—	111	139	22	.610	.191	.063	.056	.079	.099
	304	111	74	27	5	5	—	149	187	28	.610	.184	.056	.074	.070	.086
Mar. 1–7, 1954	43	17	6	2	1	1	—	14	25	4	.558	.186	.058	.174	.023	.093
10, '53	206	73	48	14	5	3	—	102	126	20	.602	.189	.049	.066	.090	.097
31, '52	37	10	6	5	—	5	2	16	24	—	.486	.162	.189	.108	.054	—
	286	100	60	21	6	9	2	132	175	24	.548	.179	.099	.116	.057	.095
Apr. 11–15, 1954	154	51	34	6	10	4	2	70	91	11	.558	.234	.052	.110	.042	.071
20, '52	78	18	20	17	2	6	4	44	54	9	.513	.205	.205	.032	.045	.115
23, '53	294	78	65	28	55	23	9	126	201	31	.480	.228	.139	.094	.058	.105
29–May 3, '56	124	32	28	4	7	10	5	52	80	14	.468	.262	.117	.113	.040	.110
	650	179	147	55	24	43	20	292	426	65	.505	.232	.128	.087	.046	.100
May 5–10, 1954	64	14	13	2	6	6	1	28	42	10	.438	.266	.109	.117	.063	.156
10, '53	171	37	23	16	5	15	6	71	122	24	.424	.213	.149	.135	.076	.140
21–24, '56	114	16	20	5	12	19	5	52	80	19	.368	.298	.162	.127	.044	.162
23, '52	20	3	2	3	—	5	1	6	16	2	.300	.225	.325	.025	.125	.100
	369	70	58	26	23	45	13	157	260	55	.383	.251	.186	.101	.077	.139

TABLE 2—*Continued*

Collection date	No. males	Numbers of zygotic combinations									Gametic frequencies					
		STST	STAR	STCH	ARAR	ARCH	CHCH	ST het.	All het.	SR	ST	AR	CH	PP	TL	SR
June 1–5, 1954	28	3	1	1	7	4	—	9	18	10	.268	.410	.107	.160	.054	.357
4–26, '53	30	3	5	1	4	6	2	11	21	5	.283	.367	.183	.117	.050	.167
14–16, '56	26	2	5	1	5	5	3	7	16	6	.231	.404	.269	.019	.038	.207
19–23, '52	70	5	17	2	4	17	9	27	51	12	.264	.329	.286	.064	.057	.171
	154	13	28	5	20	32	14	54	106	33	.262	.377	.211	.090	.050	.225
July 12–18, 1954	84	32	17	2	3	4	1	34	48	4	.583	.196	.065	.119	.036	.048
13–22, '55	96	30	12	8	2	5	9	37	54	12	.511	.147	.184	.105	.053	.126
18–19, '56	53	14	11	4	1	8	4	21	34	3	.462	.217	.208	.047	.066	.056
12–18, '56	114	33	16	13	4	14	5	45	72	15	.487	.175	.202	.061	.075	.132
	346	109	56	27	10	31	19	137	208	34	.511	.184	.165	.083	.057	.091
Aug. 1–6, 1955	48	17	11	6	2	5	—	22	28	4	.583	.219	.115	.042	.042	.083
11–15, '56	54	17	12	6	—	5	1	27	36	4	.565	.167	.148	.074	.046	.071
12–18, '54	205	67	39	23	7	7	3	99	128	11	.568	.176	.102	.115	.034	.054
	307	101	62	35	9	17	4	148	192	19	.572	.187	.122	.077	.041	.069
Sept. 3–7, 1955	73	20	11	20	3	4	2	40	48	9	.548	.158	.205	.055	.034	.123
1–12, '52	52	20	12	6	1	3	3	20	26	5	.577	.192	.144	.048	.038	.096
12–16, '54	53	19	5	8	1	1	11	19	22	*	.538	.085	.302	.075	—	*
14–18, '56	28	5	4	4	1	4	—	15	22	3	.446	.196	.179	.125	.054	.107
	206	64	32	38	6	12	16	94	118	14	.527	.158	.208	.076	.031	.109
Oct. 11, 1955	125	48	24	12	11	6	1	47	65	17	.572	.220	.112	.060	.063	.136
Nov. 1–20, 1953	90	47	14	1	2	1	—	32	41	1	.700	.135	.017	.117	.028	.011
	2851	845	556	250	116	201	89	1251	1789	293	.521	.198	.132	.100	.051	.116

* Sample not obtained.

TABLE 3. *Samples taken at Vandeventer Flat, 1952–1956*

Collection date	No. males	Numbers of zygotic combinations									Gametic frequencies					
		STST	STAR	STCH	ARAR	ARCH	CHCH	ST het.	All het.	SR	ST	AR	CH	PP	TL	SR
Feb. 10, 1953	238	70	45	25	3	9	1	125	163	18	.557	.174	.082	.084	.103	.076
Mar. 15, 16, 1953	137	22	35	10	3	7	2	72	109	14	.423	.241	.109	.117	.106	.102
27, '53	221	63	34	26	3	6	9	110	144	18	.534	.147	.122	.122	.075	.081
28–30, '56	112	26	11	13	10	12	3	45	72	18	.433	.237	.152	.134	.045	.158
	470	111	80	49	16	25	14	227	325	50	.463	.208	.128	.124	.075	.114
Apr. 10–15, 1954	152	44	38	9	5	9	3	72	100	7	.526	.230	.095	.105	.036	.046
20, '52	84	10	21	20	2	8	4	49	68	20	.411	.226	.244	.071	.048	.238
24, '53	321	101	57	40	3	17	3	145	214	22	.540	.171	.131	.089	.056	.069
	557	155	116	69	10	34	10	266	382	49	.492	.209	.157	.088	.047	.118
May 12–15, 1956	56	8	10	4	3	5	1	25	44	10	.366	.254	.145	.179	.054	.175
13, '53	182	45	37	23	8	15	4	84	124	26	.478	.228	.151	.099	.041	.143
15–17, '54	119	22	26	8	10	20	3	47	84	23	.382	.324	.168	.088	.025	.193
23, '52	150	27	24	25	9	27	13	61	100	37	.383	.247	.283	.063	.023	.247
	507	102	97	60	30	67	21	217	352	96	.402	.263	.187	.107	.036	.190
June 1–5, 1954	74	12	6	8	8	11	4	24	48	20	.324	.277	.209	.122	.061	.270
4, '55	183	55	28	16	2	11	—	85	124	42	.533	.167	.098	.139	.060	.230
17, '53	66	12	14	4	5	7	5	25	44	17	.371	.280	.205	.106	.038	.258
	323	79	48	28	15	29	9	134	216	79	.409	.241	.171	.122	.053	.253
June 24, 1952	35	6	9	4	3	6	—	15	26	5	.386	.343	.157	.086	.029	.143
26, '53	289	71	60	29	14	26	16	129	187	51	.469	.216	.178	.087	.043	.176
29, '53	81	17	19	9	1	12	2	38	61	11	.444	.247	.179	.099	.031	.136
27–29, '56	181	40	37	22	17	26	8	62	115	33	.420	.301	.188	.055	.036	.177
	586	134	125	64	35	70	26	244	389	100	.430	.277	.176	.082	.035	.158

TABLE 3—*Continued*

Collection date	No. males	Numbers of zygotic combinations									Gametic frequencies					
		STST	STAR	STCH	ARAR	ARCH	CHCH	ST het.	All het.	SR	ST	AR	CH	PP	TL	SR
July 4–10, 1954	115	24	30	9	10	15	3	52	78	13	.435	.291	.170	.078	.026	.113
5–8, '55	115	21	14	11	4	15	11	54	79	18	.417	.165	.243	.108	.065	.157
19–24, '56	122	26	24	20	10	20	4	52	82	11	.426	.279	.221	.049	.025	.090
28, '52	75	29	18	11	3	7	1	32	42	7	.600	.227	.133	.027	.013	.093
	427	100	86	51	27	57	19	190	281	49	.470	.241	.192	.066	.032	.113
Aug. 12–18, 1954	89	20	21	13	4	14	3	40	62	15	.449	.275	.197	.062	.017	.169
15, '53	124	38	25	21	16	11	2	51	68	21	.512	.294	.149	.032	.012	.169
23–25, '55	94	19	22	24	1	12	6	51	68	11	.473	.202	.271	.037	.016	.117
24–27, '56	108	22	18	7	10	24	15	30	61	15	.343	.306	.296	.023	.032	.138
	415	99	86	65	31	61	26	172	259	62	.444	.269	.228	.039	.019	.148
Sept. 12–16, 1954	157	41	30	25	5	20	10	66	101	21	.471	.207	.239	.051	.032	.134
23–26, '56	61	14	7	10	4	7	2	23	41	10	.418	.205	.238	.074	.066	.164
26, '53	141	50	28	9	6	4	3	55	80	18	.550	.209	.085	.082	.074	.128
	359	105	65	44	15	31	15	144	132	49	.480	.207	.187	.070	.057	.142
Oct. 1–3, 1955	142	17	20	22	5	14	18	48	100	34	.289	.197	.345	.109	.060	.239
13–28, '53	35	8	7	4	2	—	—	14	25	7	.429	.214	.143	.089	.129	.200
	177	25	27	26	7	14	18	62	125	41	.359	.206	.244	.099	.095	.220
	4059	980	775	481	189	397	159	1781	2624	593	.451	.230	.175	.088	.055	.153

TABLE 4. *Samples taken at Keen Camp, 1952–1956*

Collection date	No. males	Numbers of zygotic combinations									Gametic frequencies					
		STST	STAR	STCH	ARAR	ARCH	CHCH	ST het.	All het.	SR	ST	AR	CH	PP	TL	SR
Mar. 7, 1954	54	13	5	7	5	5	1	18	34	1	.407	.250	.167	.120	.056	.018
12–21, '56	37	7	3	7	3	10	2	12	24	2	.351	.270	.297	.081	—	.048
	91	20	8	14	8	15	3	30	58	3	.379	.260	.232	.100	.028	.033
May 1, 1952	112	15	26	26	1	4	2	66	93	15	.429	.196	.196	.094	.085	.133
1–3, '56	72	7	17	3	4	12	5	32	56	11	.320	.313	.194	.111	.063	.149
1–5, '54	149	21	34	12	11	25	7	60	109	19	.342	.322	.198	.097	.037	.127
6, '53	213	36	45	32	15	19	9	95	151	23	.392	.272	.195	.099	.035	.107
	546	79	122	73	31	60	23	253	409	68	.370	.276	.196	.100	.055	.129
June 1, 1953	25	7	7	3	1	1	—	10	17	*	.480	.280	.100	.080	.060	*
1–4, '56	143	13	30	24	11	24	18	65	100	13	.318	.287	.311	.059	.024	.096
3, '52	41	3	10	7	4	5	6	21	28	5	.329	.305	.293	.037	.037	.121
6, '55	113	12	29	27	3	19	5	62	93	13	.381	.265	.270	.049	.035	.115
	322	35	76	61	19	49	29	158	238	41	.377	.284	.244	.056	.039	.111
June 23–26, 1954	111	13	21	14	7	26	9	45	81	*	.320	.284	.297	.081	.018	*
27–30, '56	178	23	26	33	7	45	16	72	132	11	.331	.264	.323	.053	.028	.061
July 1–4, 1953	96	16	26	16	3	8	5	49	72	10	.422	.255	.208	.073	.042	.104
4–6, '52	90	10	16	19	3	18	16	39	61	19	.328	.233	.394	.022	.022	.208
	475	62	89	82	20	97	46	205	346	40	.350	.259	.306	.057	.028	.125
July 23–27, 1954	129	15	35	22	6	26	8	61	98	9	.353	.302	.271	.050	.023	.069
29–Aug. 2, '56	61	7	16	15	4	10	9	25	41	9	.320	.221	.369	.074	.016	.145
Aug. 8–11, 1952	31	4	6	6	2	6	3	12	22	4	.323	.290	.323	.032	.032	.129
12–15, '55	165	24	35	34	5	35	12	79	124	25	.385	.261	.294	.036	.024	.152
	386	50	92	77	17	77	32	177	285	47	.345	.269	.314	.048	.024	.124
Aug. 28–31, 1954	139	23	24	25	4	19	10	61	101	14	.385	.216	.273	.079	.043	.100
Sept. 4–6, 1956	63	1	7	18	1	20	11	26	51	9	.222	.238	.500	.024	.016	.143
15, '53	43	5	7	7	2	2	4	23	31	5	.384	.186	.233	.140	.047	.116
21–25, '55	87	11	10	18	—	19	18	33	58	21	.316	.172	.448	.034	.029	.241
24–28, '54	187	14	44	41	14	33	20	97	139	32	.334	.286	.321	.032	.027	.171
	519	54	92	109	21	93	63	240	380	81	.328	.220	.355	.062	.032	.154
	2339	300	479	416	116	391	196	1063	1716	280	.358	.261	.274	.071	.034	.113

* Sample not obtained.

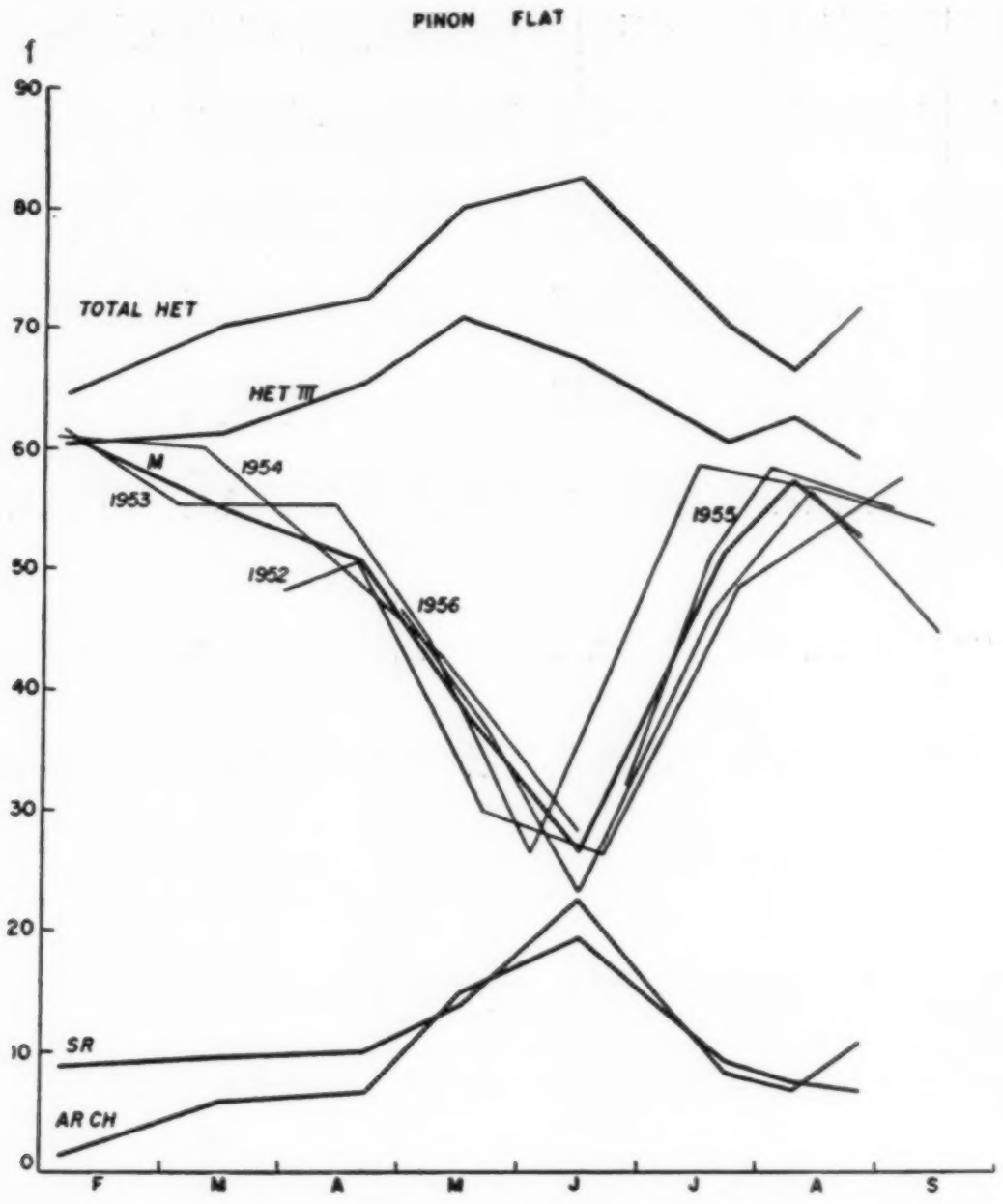

FIG. 1. Changes in the frequency system during the seasonal rhythm at Pinon Flat, referred to in the text and abstracted from table 2.

an almost identical pattern, as might be expected. This rhythm has been remarkably regular each year, relative to those of AR and CH, suggesting that the frequencies of ST are somehow primarily concerned with the rhythm. As ST changes in frequency, AR and CH change inversely but not equally in each case, AR being more frequent in some years than CH, and vice versa. Their relative frequencies in 1952, for example, were respectively, .215 and .225; in 1953, .220 and .100; in 1954, .200 and .110; in 1955, .186 and .154; and in 1956, .257 and .181. The rhythm at Vandeventer Flat has been similar but the change in frequencies has been less pronounced and less regular than at Pinon Flat. Observation suggests that part of the irregularity can be ascribed to a greater variation in earliness and lateness of spring and summer, depending on the relative influence of desert and coastal conditions of weather at this station each year. At Keen Camp, no regular change is perceptible.

Turning now from changes in gametic frequency to those of the surviving male zygotes we find a seasonal change in heterozygotes that is both qualitative and quantitative. The shift in ST gametic frequencies is accompanied by a similar shift in the principal ST heterozygotes, both STAR and STCH decreasing and increasing concurrently with ST gametes

but not at the same rates (table 1). The principal heterozygote of the third chromosome at both stations to change inversely to the frequency of ST is ARCH. Its rhythm is shown at the bottom of each figure. Its increase at Pinon Flat is sufficient to increase the total heterozygosity of the third chromosome to a maximum in May and June, as shown in the upper range in figure 1. Total third chromosome heterozygosity does not apparently increase at Vandeventer Flat in spite of a similar increase of ARCH, but, again, differences in season may have obscured an increase on the chart. No regular changes of either gametic or zygotic frequencies have been found at Keen Camp.

Associated with the increase of ARCH and other heterozygotes of the third chromosome present in lower frequencies, is a corresponding increase of the SR arrangement of the X chromosome. The over-all increase of heterozygosity in both chromosomes that results therefrom during the seasonal rhythm has been computed and is shown in the topmost curve of each figure. This curve is taken from table 5 which has been designed to estimate these trends of frequency in the females (to which crossing over is restricted). We have already observed

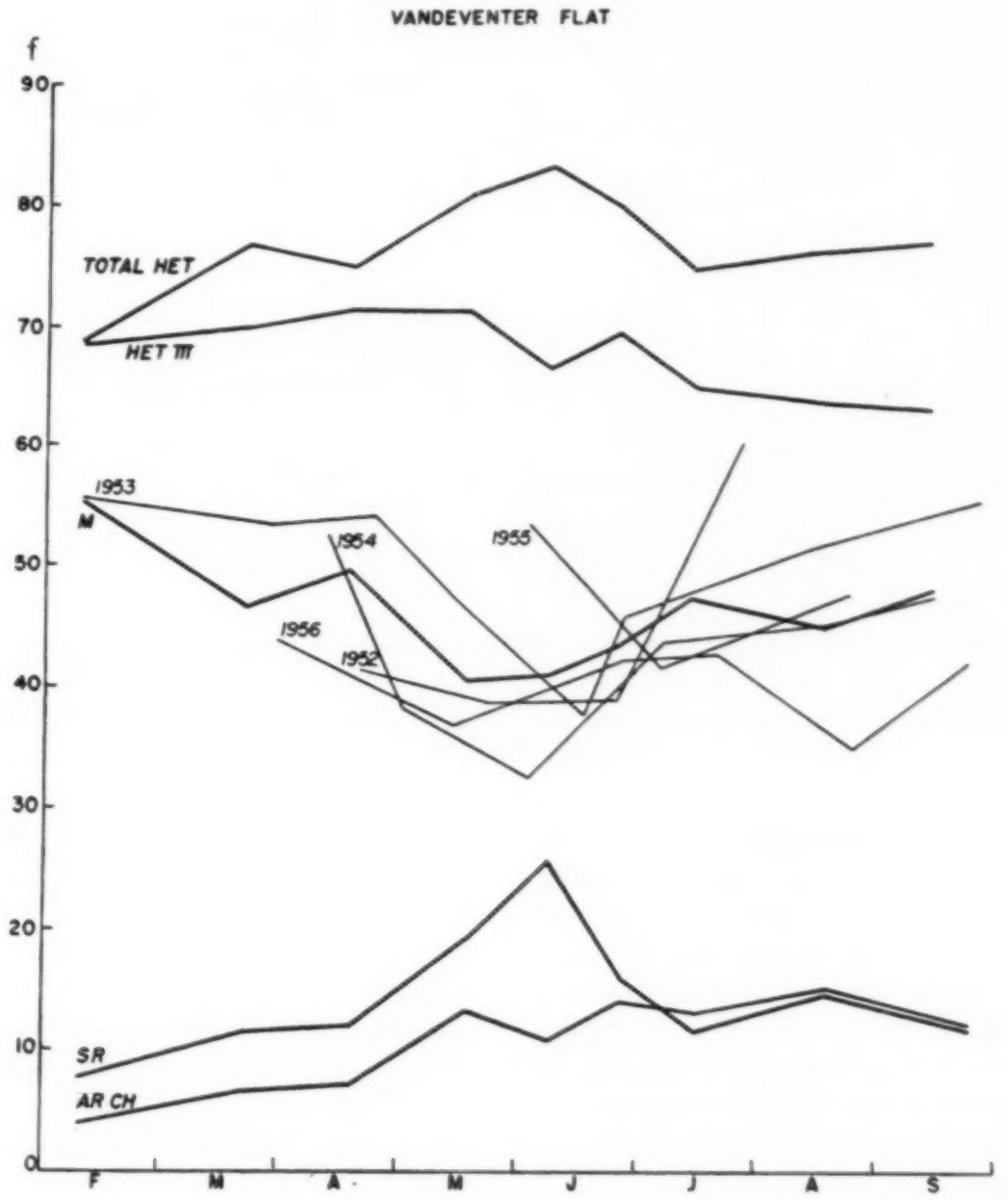

FIG. 2. Changes in the frequency system during the seasonal rhythm at Vandeventer Flat referred to in the text and abstracted from table 3.

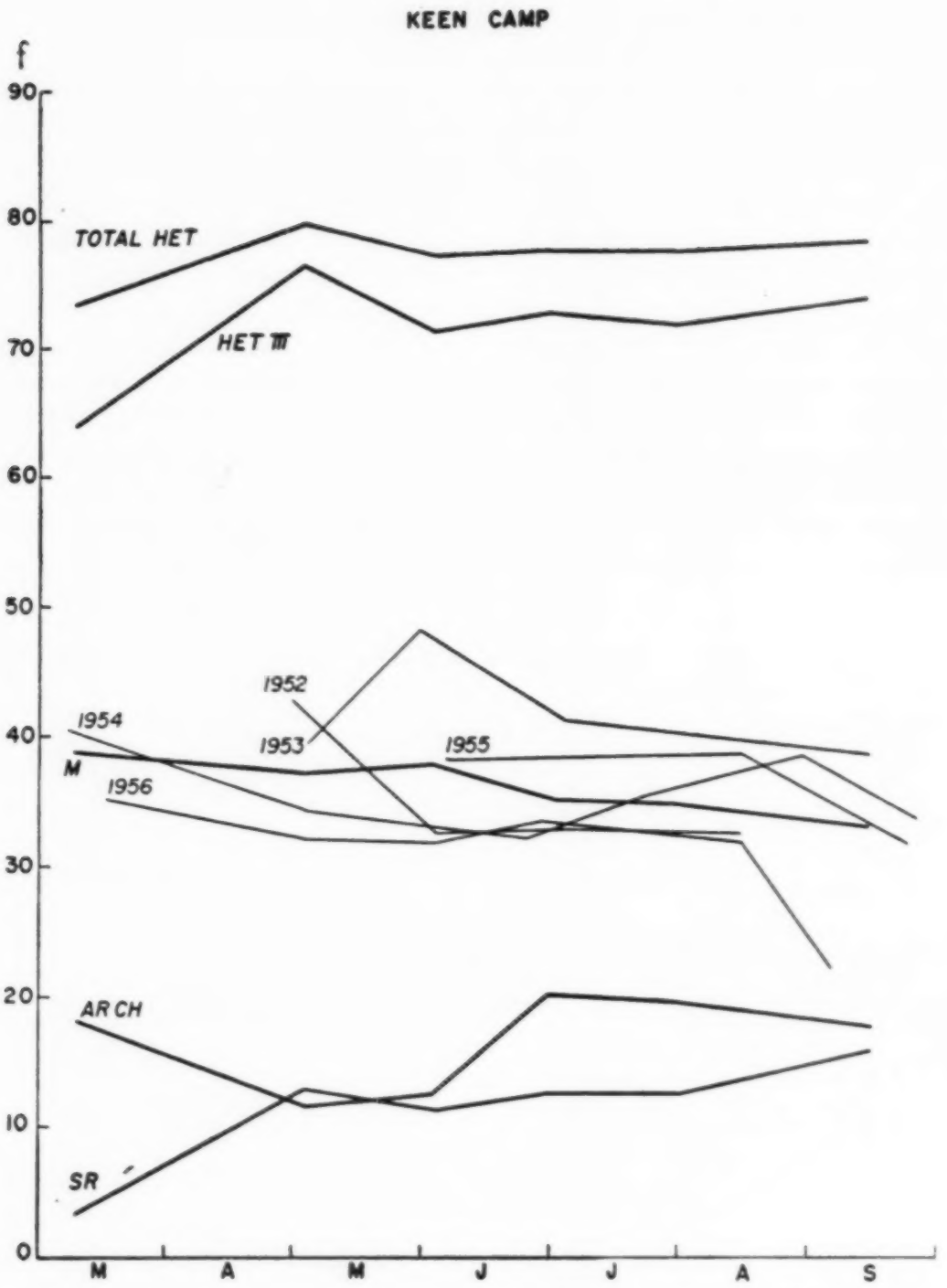

FIG. 3. Changes in the frequency system at Keen Kamp referred to in the text and abstracted from table 4.

that the actual frequencies may differ between the sexes, but that the trends of frequency in both chromosomes are similar. Present evidence also indicates that the relative frequencies of homozygotes and heterozygotes in each sample do not deviate significantly from the expected values of the binomial square rule. We have therefore computed the values for heterozygous females shown in table 5 by the formula $1 - (p^2 + q^2 + r^2 + s^2 + t^2)$; in which p is the frequency of ST, q that of AR, etc. These values appear in line A of table 5. The frequencies of the heterozygous females in the X chromosome were computed by the formula $2q\ (1 - q)$ in which the frequency of the SR arrangement in the males was taken as q and that of STX as $1 - q$. These values appear in line B of the table. Taking these estimates, we then estimated the proportion of females likely to be simultaneously heterozygous for both chromosomes by multiplying the two values. These products

TABLE 5. *Mean monthly frequencies of inversion heterozygotes during 1952–1956, estimated for females*

	Pinon Flat								Vandeventer Flat									Keen Camp						
	Feb.	Mar.	Apr.	May	June	July	Aug.	Sept.	Feb.	Mar.	Apr.	May	June	June	July	Aug.	Sept.	Mar.	Apr.	May	June	July	Aug.	Sept
	Expected frequencies of third chromosome heterozygotes																							
A	.581	.640	.665	.740	.734	.668	.615	.647	.635	.705	.680	.722	.728	.699	.679	.677	.692	.724		.735	.713	.713	.707	.713
	Expected frequencies of X chromosome heterozygote																							
B	.157	.172	.180	.239	.349	.165	.128	.194	.140	.202	.208	.308	.378	.266	.200	.253	.244	.064		.225	.197	.217	.217	.261
	Expected frequencies of females heterozygous for both chromosomes																							
C	.091	.110	.120	.177	.256	.110	.079	.126	.089	.142	.141	.222	.275	.186	.136	.171	.169	.053		.165	.140	.155	.153	.186
	Expected frequencies of females heterozygous only for the third chromosome																							
D	.490	.530	.545	.563	.478	.558	.536	.521	.546	.563	.539	.500	.453	.513	.543	.506	.523	.671		.570	.573	.558	.554	.527
	Expected frequencies of females heterozygous only for the X chromosome																							
E	.066	.062	.060	.062	.093	.055	.049	.068	.051	.060	.067	.086	.103	.080	.064	.082	.075	.011		.060	.057	.062	.064	.075
	Estimated over-all heterozygosity of females																							
F	.647	.702	.725	.802	.827	.723	.664	.715	.686	.765	.747	.808	.831	.779	.743	.759	.767	.735		.795	.770	.775	.771	.778

appear in line C. Then, by subtracting the values of line C from line A, we got an estimate of heterozygosity in the third chromosome only, which is shown in line D. Similarly, by subtracting the values of line C from line B, we got an estimate of heterozygosity only in the X chromosome, which is shown in line E. The final estimate of over-all heterozygosity in each chromosome and both simultaneously was made by adding the values of lines C, D and E. These values appear in line F and are those graphed in figures 1, 2 and 3.

In sum, then, the seasonal rhythm of frequencies has two aspects. Over-all structural heterozygosity in about 40 per cent of the euchromatin increases and then decreases regularly. Second, the rhythm is accompanied by regular changes in the principal homozygous classes, STST being replaced by ARAR and CHCH, and then regaining its former frequency.

In view of the strong selection that takes place during the rhythm it is surprising to find no indication of it in the differential survival of zygotic combinations. But this seems to be the case. The summed deviations by year of the principal combinations in each monthly sample at Pinon Flat, and the yearly summations, are diagrammed in Figure 4, in order to indicate their regularity (see also Epling, Mitchell and Mattoni, 1955). The totals for the whole period are given in table 6. The data show that STST and CHCH have been in excess in most samples and that ARAR has been deficient. This is not in accord with the decrease of STST and the increase of ARAR and CHCH during the rhythm. It may be, however, that selection is operating on other attributes that do not affect the Hardy-Weinberg ratios, such as relative fecundity, or because of interchromosomal effects on the genotypes produced each generation.

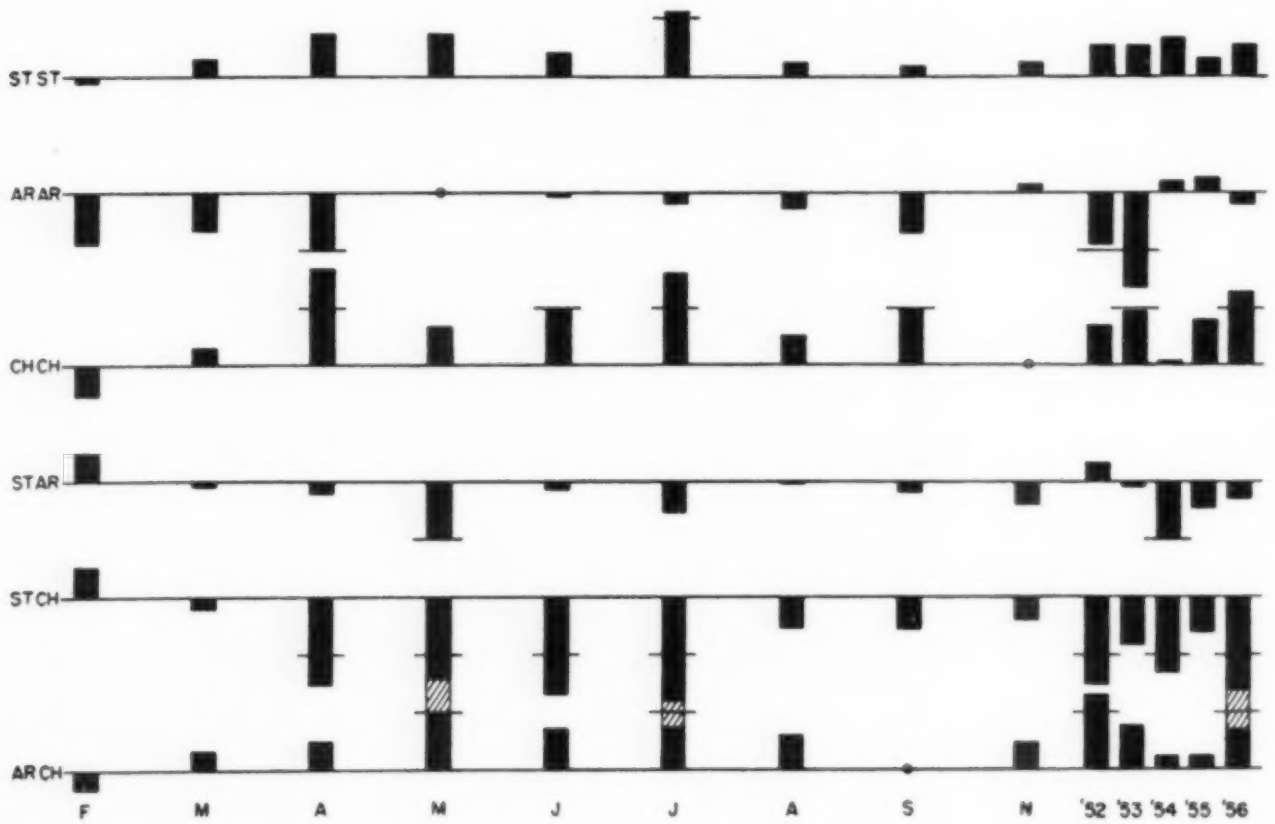

FIG. 4. Deviations of each of the principal zygotic combinations at Pinon Flat from the values expected of the binomial square rule, based on the data given in table 2. Each bar on the left represents the deviation of the summed monthly samples. The bars on the right are the summed samples for each year. The short horizontal lines indicate a significance level of 5%. The cross hatching simply indicates areas of overlap of certain bars (see also Epling, Mitchell and Mattoni, 1955).

TABLE 6. *Yearly summations of the deviations from the binomial square rule of the zygotic combinations indicated*

	Pinon Flat											
	STST		ARAR		CHCH		STAR		STCH		ARCH	
	σ^2	x−e	σ^2	x−e	σ^2	x−e	σ^2	x−e	σ^2	x−e	σ^2	x−e
1952	6.162	+ 8.70	17.028	− 7.24	17.076	+ 5.83	55.324	+ 4.98	55.828	−22.73	31.519	+14.16
1953	201.396	+15.70	41.775	−21.13	11.454	+ 6.53	169.682	− 2.72	84.053	−15.25	40.114	+ 9.81
1954	147.656	+15.99	29.445	+ 6.25	6.919	+ .93	119.865	−22.98	63.140	−19.98	23.793	+ 2.17
1955	44.42	+ 3.99	5.96	+ .83	6.67	+ 4.08	31.70	− 5.14	32.81	− 6.73	11.59	+ 1.70
1956	56.49	+ 8.48	24.56	− 1.90	9.67	+ 7.96	63.18	− 5.08	41.71	−29.63	28.40	+15.28
Total	510.124	+52.86	118.768	−23.19	51.789	+25.33	439.751	−30.94	277.541	−94.32	135.416	+43.12
$\frac{x-e}{2\sigma}$	+1.17		−1.06		+1.76		−.738		−2.83		+1.85	

DISCUSSION

The foregoing data suggest to us that there are four principal aspects of the seasonal rhythm to be explained:

1. The regularity of the rhythm at Pinon Flat, its irregularity at Vandeventer Flat, and its absence at Keen Camp.
2. The greater regularity of ST during the rhythm than AR and CH.
3. The restitution of the original system of frequencies during the rhythm rather than the continuance of a new system once it has become established.
4. The increase of heterozygosity during the rhythm when at Pinon Flat it results in an increase in both chromosomes and at Vandeventer Flat an increase only in the X chromosome.

We base our explanation primarily on interchromosomal interactions on crossing over and recombination. It rests on the following propositions.

1. The gene arrangements of a given chromosome are genetically and adaptively different. The observed regularities of ecogeographical distribution are strong presumptive evidence of this. These differences are preserved by the restrictions on crossing over in structural heterozygotes (Sturtevant and Beadle, 1936; Dobzhansky and Epling, 1948). The consequences of this restriction underlie the theory of balanced chromosomal polymorphism which has been discussed in numerous papers, particularly by Dobzhansky.

2. The genome is an operative unit in respect of crossing over. That inversion heterozygotes have complex and variable interchromosomal effects on crossing over is well documented (Morgan, Bridges and Schultz, 1933; Steinberg, 1936, 1937; Komai and Takaku, 1940, 1942; Steinberg and Fraser, 1944; Schultz and Redfield, 1951; Carson 1953; Levine and Levine 1954, 1955). We assume that total crossing over remains more or less constant (Mather, 1936) and that different degrees and kinds of structural het-

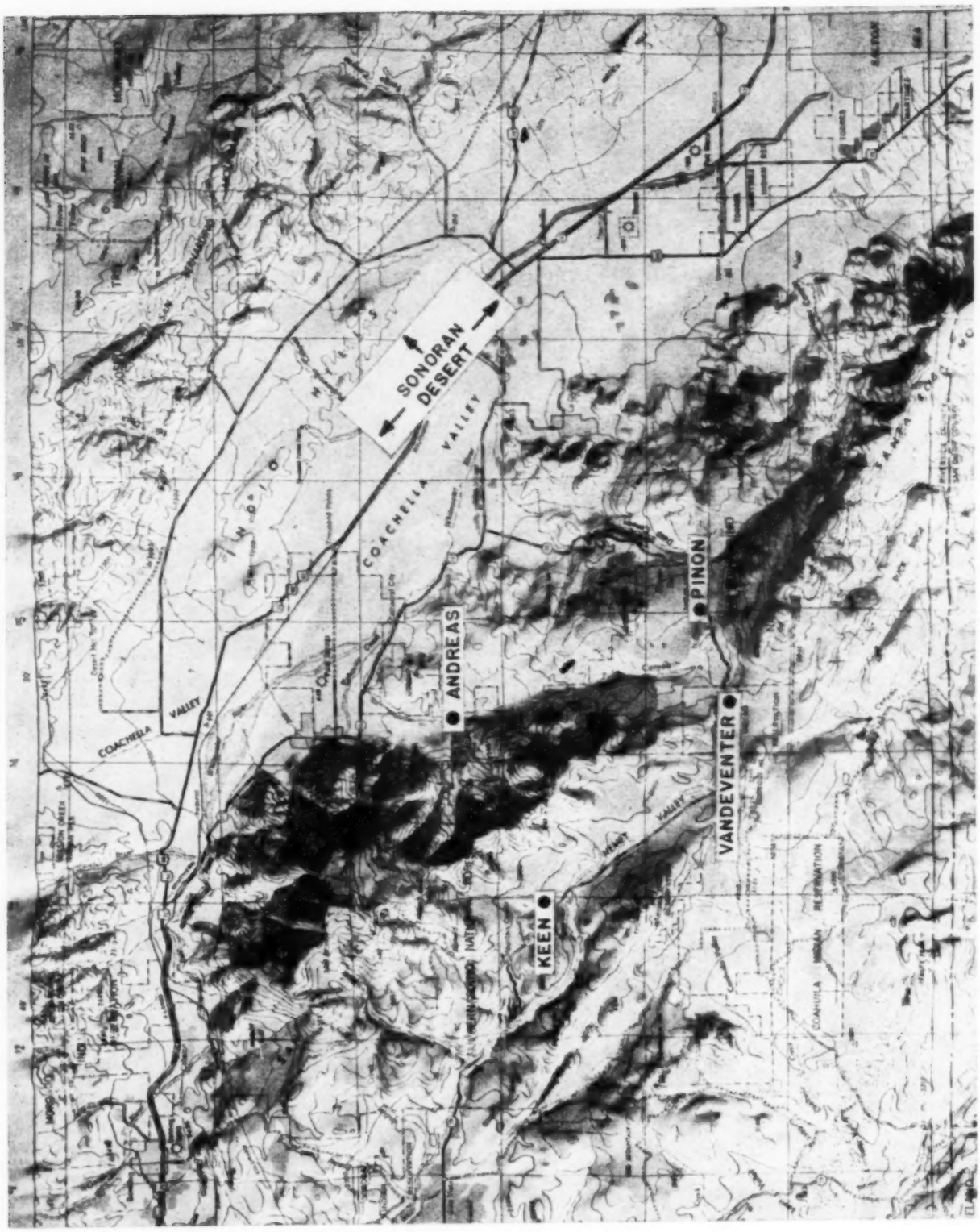

FIG. 5. Photograph of an embossed topographic map of the San Jacinto and Santa Rosa ranges, showing the relative positions of the collecting stations referred to. The reader will note that Pinon Flat and Vandeventer Flat are situated at about the same elevations in the saddle between the two ranges. The barrier these ranges make is suggested by the shadows that would be cast by the afternoon sun.

erozygosity may alter the patterns of crossing over and recombination in wild populations.

3. A certain pattern of relative frequencies of third chromosome arrangements is normally adaptive to the mediterranean climate of the Pacific slope (figures on pp. 127, 129, Dobzhansky and Epling, 1944).

4. This pattern is adaptively qualified by regularly increasing frequencies southward of the sex ratio arrangement of the X chromosome.

5. The resulting increase of multiple heterozygosity is accompanied by release of variability (Mitchell, unpublished data on *D. melanogaster*).

The stations referred to are situated along an environmental gradient from Pinon Flat (and Andreas Canyon as shown earlier by Dobzhansky) to Keen Camp. In view of the seasonal differences of response in the populations along this gradient, it is appropriate now to describe it. The collecting area is about 125 miles east of Los Angeles (fig. 5). It is dominated by two mountain blocks, the San Jacinto and the connected Santa Rosa range which together interpose a barrier between the coastal plain and the coastal aspect of the mountains themselves and the western margin of the Sonoran desert. Mount San Jacinto rises to an elevation of 10,381 feet; Toro Peak, or Santa Rosa Mountain, rises to 8716 feet. The lowest elevation between them is about 4500 feet. The desert floor which lies about 5–10 miles north and east has an elevation of 500 feet or less. The altitudinal gradient is so abrupt that a difference of 10,000 feet exists at some places over a map distance of about seven miles. The environmental gradient is correspondingly abrupt and the extremes of environment are equivalent to latitudinal differences in the species area from the Mexican border to British Columbia. These differences are reflected in the vegetation. Intermediate elevations on the coastal side are occupied by a forest like that in the Sierra Nevada at Mather (Dobzhansky, 1948). Both are dominated by the same species, have many species in common, and indicate similar environmental conditions. A subalpine forest is also found at higher elevations but more impoverished than that at the Timberline station of Dobzhansky (1948; Clausen, Keck and Heisey, 1940). Below the yellow pine forest, as elsewhere in California, is an altitudinal zone of chaparral, a brushland dominated by the same or related species in most of its range in California. The population density of *D. pseudoobscura* is generally greater in the forest than in the chaparral, which is greater than that in the brushland of the desert. This coastal facies, one will remember, is controlled by a mediterranean climate in which precipitation generally reaches its peak during the coldest months, after which a protracted drought follows during the warmest part of the year. Summer showers are largely confined to higher elevations, and particularly to slopes looking toward the interior.

The interior slopes are very different. The forest at higher elevations quickly gives way to a zone of chaparral or passes directly into a zone of pinon-juniper. These are soon succeeded by the shrubby and annual genera that range so widely over the Sonoran desert and its bordering ranges. The climatic regime is that of the Intermontane Plateau, a composite or mosaic of the mediterranean climate of the Pacific slope and that of the Sonoran desert. Precipitation is diminished altitudinally and the temperatures are relatively high, with a corresponding saturation deficit (Figure 2 on page 157 of Dobzhansky and Epling, 1944).

The four collecting stations to which we shall refer form a quadrangle along this gradient. Keen Camp lies on the slope of Mt. San Jacinto at about 4500 feet elevation in the lower reaches of the yellow pine forest and chiefly within the influence of the coastal climate. The veg-

FIG. 5. Photograph of an embossed topographic map of the San Jacinto and Santa Rosa ranges, showing the relative positions of the collecting stations referred to. The reader will note that Pinon Flat and Vandeventer Flat are situated at about the same elevations in the saddle between the two ranges. The barrier these ranges make is suggested by the shadows that would be cast by the afternoon sun.

etation and the temperature and rainfall records (Epling, Mitchell and Mattoni, 1953) indicate that the environment is not dissimilar to that at intermediate elevations in the central Sierra Nevada. Vandeventer Flat lies at about the same elevation in the gap between the two mountain ranges in a local and marginal area of chaparral, that is, in coastal vegetation which is ecologically transitional to the shrubby communities of the desert. The station is situated in a small grove of ancient liveoaks which are supported by an intermittent stream. The oak, *Quercus agrifolia,* and the community itself, both of which are typically coastal, reach their ecological limits at this point, probably because of the influence of desert temperatures and rainfall like those at Pinon Flat which is only six miles away. Pinon Flat is at an elevation of 4000 feet in the gap but on the desert slope of Santa Rosa mountain in an open woodland of pinon pine and desert juniper, intermixed with many desert shrubs. This community extends many miles eastward in one guise or another along the bordering ranges of the Sonoran desert. In winter and spring its climate is influenced more by the rainfall and temperatures of the coast than those of the interior. In summer the desert temperatures are predominant, and are accompanied by summer showers in some years. Its climate, of any of the stations, is most like that of Arizona where AR and SR reach their greatest frequencies. Andreas Canyon, for which only Dobzhansky's data (1943) are available for comparison, is at an elevation of about 800 feet at the base of the mountains along the margin of a living stream. Its climate is that of the desert floor except as far as it is modified by the stream and its woody vegetation.

Two particulars of this gradient should be noted. It is situated along an interface between two climatic regimes and also at an interface between the ST system of the Pacific slope and the AR system of the Sonoran desert. The environmental consequences of the climatic interface are as follows. The winter rains which are a feature of the mediterranean coastal climate occur simultaneously at Keen Camp and at Pinon Flat, but total precipitation is much less at the latter. It is often in the form of snow at Keen Camp which results in less runoff and a greater use by the vegetation of the water available. The onset of warm weather is much earlier at Pinon Flat which is more influenced by desert temperatures. It follows on the heels of the winter rains and induces an earlier growing season of the vegetation on which Drosophila depends. Desiccation follows as much as two months earlier than at Keen Camp and is more drastic because of the lower precipitation and higher temperatures. Winter and spring are telescoped into a much shorter period at Pinon Flat and the result is a rapid shift in the biotic cycle which lessens along the gradient toward Keen Camp where the seasonal rhythm approaches the norm for the yellow pine forest in California. In summer the differences of weather are less marked between these stations and both sites enter a period of summer dormancy during which changes are more gradual and the mediterranean climate of the coast finally becomes uppermost in influence at both stations.

The relation of these changes of environment at Pinon Flat to the seasonal rhythm of frequencies is illustrated in figure 6. The seasonal rhythm of weather at Pinon Flat is indicated in this figure by daily changes of temperature, registered as total heat input above forty degrees mean temperature, and the more or less parallel saturation deficit. These data were obtained in 1951–1952 from a continuous daily record registered in a hygrothermograph. The derivation of the two factors is described in Epling, Mitchell and Mattoni (1953). The pattern at Keen Camp is similar but the development of higher temperatures and drought during spring lag behind those of Pinon Flat until summer, when they become

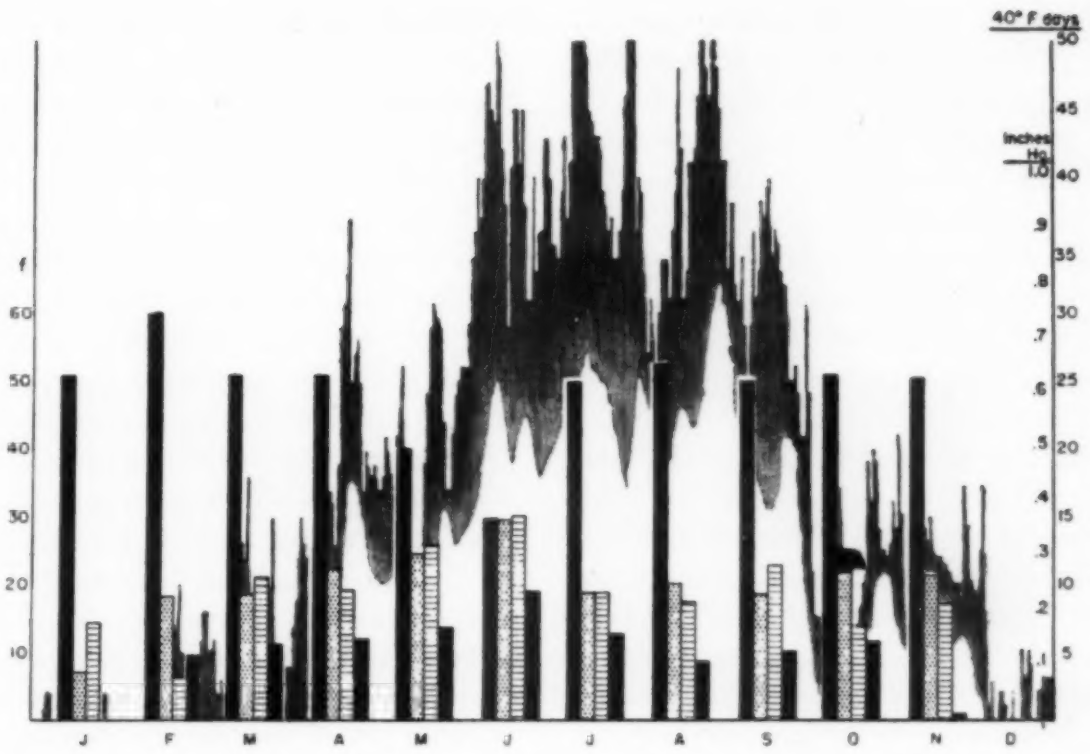

Fig. 6. The mean monthly frequencies of ST (black), AR (dotted), CH (hatched) and SR (black) at Pinon Flat since 1939, against a background that represents the seasonal rhythm of weather during 1951–1952 registered on a hygrothermograph. The uppermost curve registers the relative heat input, the lower, the saturation deficit. The derivation of the background data is described in Epling, Mitchell and Mattoni (1953).

more or less parallel at both stations. The frequencies of ST, AR, CH and SR in figure 5 represent the means of monthly collections since 1939. They vary from year to year, as we have pointed out, ST being much more consistent in its changes from year to year than AR and CH. We should like to recall also at this point, the relatively uniform mean frequency of ST along the Pacific slope, suggesting that its function in the whole population is somehow related to the general aspects of the mediterranean climate of the coastal area. One will recall, also that SR diminishes northward, as one departs from the conditions that characterize the interior desert.

The differences of weather along the environmental gradient have noteworthy effects on seasonal population densities. These can be estimated only from the ease or difficulty with which collections can be made, but prolonged experience since 1939 indicates that the following observations are probably valid. The maximum density at Pinon Flat occurs generally in March or April, or perhaps even in February, depending on the earliness or lateness of the season each year and the severity of the winter. Thereafter, it declines rapidly to a minimum in June, when collections are difficult to make and samples are small. Yet, even though the weather becomes warmer and drier in July and August, the population density increases in most years (table 2). Thus, the rhythm of ST at Pinon Flat (fig. 6) is more or less paralleled by changes in population density, with the proviso that this density is by far the highest in early spring and less in summer and autumn. Collections at Vandeventer Flat are more readily obtained in summer than at Pinon Flat and the population density is probably higher, in general, than at Pinon Flat. Nevertheless it appears to decline in some years correlatively with the decline at Pinon Flat. The population at Keen Camp reaches its maximum in July or August, which is the norm elsewhere along the Pacific slope.

Bearing these data in mind, we now offer an hypothesis to account for them. We shall illustrate it by an explanation

of what occurs each year during the seasonal rhythm of weather at Pinon Flat, alluding later to the differences of the rhythm at Vandeventer Flat and Keen Camp. This explanation is doubtless oversimplified because many variables are involved. It does not answer every question. But we believe that it outlines the general sequence of adaptive events at each station and that it can be tested by experiment.

The data presented in the earlier part of the paper suggest to us that two kinds of interchromosomal relations need be taken into account to explain the seasonal rhythm. The first of these is a quantitative relation in respect of crossing over in each chromosome. If we assume that total crossing over in the genome is maintained at a fairly steady state, as Mather (1936) has argued, an increase of structural heterozygosity in the third and X chromosomes would be followed by an increase of crossing over in the structurally homozygous second and fourth chromosomes. This increase may be differentially localized if the observations of Schultz and Redfield (1951) on multiple heterozygosity in *D. melanogaster* apply to *D. pseudoobscura.* And, if we regard the genome as an operative unit, and if certain crossover products in the second and fourth chromosomes should confer an adaptive advantage on a genotype, we can suppose, conversely, that the progeny of females heterozygous for both the third and X chromosomes would be selected, and over-all heterozygosity would increase. This would not of itself account for the regular increase of ARCH each year, nor for the fact that the former system of frequencies is restored. We suggest therefore that a qualitative relation exists between certain arrangements that results in their coadaptedness to some conditions but not to others.

The ecogeographical clines of relative frequency of the different arrangements indicate that each arrangement is genetically and adaptively different. The more complex cline of association between the principal arrangements of the third chromosome and SR, and the variable but regularly changing frequency systems that result, suggest a closer adaptive relation between AR and SR than between ST and SR. The uniformity of the frequencies of ST, AR and CH in the mediterranean climate along the Pacific slope suggest that the influence of these frequencies on recombination, in conjunction with STX, is effective for maintaining the populations in these conditions. We find, however, that SR increases southward in California as the climate becomes progressively influenced by that of the Sonoran desert, where SR and AR reach their maximum association. The persistence at the three stations in the San Jacinto Mountains of the mean relations between ST, AR and CH on one hand, and SR on the other, which is illustrated in table 1, suggests that the interplay in these frequency systems is adequate to adapt each population to the normal progression of the seasonal rhythm, which is predominantly coastal in aspect. The gradient along which they are situated leads, however, to the desert margin where the environment is temporarily dominated each year by the desert climate. The result is an environmental stress at Pinon Flat for which the prevailing genetic system, dominated by ST and STX, is ineffective. The population at Vandeventer Flat is submarginal and the desert influence is variably expressed and is generally less pronounced. The population at Keen Camp is coastal, with conditions not unlike those of the Sierra Nevada. The vegetation types at each station substantiate these inferences.

We assume that interactions between certain crossover products in the second and fourth chromosomes in conjunction with those in STST and STXSTX are generally adaptive to the coastal rhythm of seasons and biotic development, and that as long as this rhythm prevails, certain frequencies of the different arrangements will be maintained. If the conditions of the stress should result in a high

selective advantage for other crossover products in the second and fourth chromosomes, and if these are adaptively associated with AR, CH and SR, rather than with ST, then selection of the progeny of females heterozygous for these arrangements would result in an increase of ARCH and STXSR, which is observed. Thus, the seasonal rhythm of frequencies can be ascribed to quantitative and qualitative changes in recombination that are mediated to the environmental changes by the inversion system present.

The release of variability through these interchromosomal actions reaches its maximum sometime in June or early July, depending partly on the season, and it must be remembered that the abrupt shift in June shown in Figure 1 is probably in part an artifact of collecting. If adequate collections could be obtained weekly during this period, the real situation may be more like that shown in the graph for 1952 when a plateau was reached for a short period. But taken on the long time average, it appears that maximum variability is reached sometime during this period when, following intense selection and the decline in population density, ST, AR and CH become equal in frequency and STX and SR approach it (fig. 6). The increase of population density observed in July or August suggests that sufficient variability has been released to accommodate the population to the conditions imposed by the stress.

When the influence of the desert climate recedes and conditions at Pinon Flat become more like those generally found in July and August in the coastal areas, the recombinants formed by crossovers in the second and fourth chromosomes that are normally associated with STST and STXSTX would be more adaptive. Selection for the progeny of females homozygous for STX would automatically reduce over-all and multiple heterozygosity, inasmuch as only two arrangements are present in the X chromosome; and the norm of interchromosomal actions would be restored.

With this explanation of the seasonal rhythm at Pinon Flat in mind, we can account for the less pronounced and more variable rhythm at Vandeventer Flat on the assumption that the seasonal stress is expressed there less regularly and with variable intensity because of its more coastal situation and the predominance of the coastal climate most of the year. Similarly, no rhythm would be expected at Keen Camp, which falls within the coastal climatic regime, as its vegetation attests.

If confirmed, our hypothesis will account for the continually varying proportions of arrangements in space and time, each of which is adaptively different because of the suppression of crossing over. Confirmation will depend on what is learned of the effects of various heterozygotes and their associations in crossing over in the homozygous chromosomes. If polygene linkages of adaptive significance to the population occur in sections of the second and fourth chromosomes in which crossing over is normally low or negligible, and if their frequencies are increased significantly by the presence of multiple heterozygotes in the third and X chromosomes, then our hypothesis will be greatly strengthened. Marker stocks are being constructed to test this assumption. The hypothesis might also be supported by field studies carried on in other critical areas such, for example, as the interface between the AR and PP systems of frequency. Meanwhile, we are inclined to regard an inversion system as a mechanism that is regulated by adaptive responses to changing or different environments. It preserves the adaptability of a species by mediating recombination. It is a mechanism that fosters a steady state of the genetic system of each locality.

Summary

The inversion system of *D. pseudoobscura* is effectively confined to the third and X chromosomes. The suppression of crossing over in the heterozygotes of these chromosomes permits the develop-

ment of genetically different gene arrangements. The ecogeographical clines of the principal arrangements suggest that each makes a different contribution to the adaptedness of the populations in which they occur. The ecogeographical clines of relative frequencies of the arrangements in both chromosomes suggest, further, that the adaptedness of a population depends not simply on the aggregate of arrangements present but also on their interchromosomal effects. These effects can be advantageously studied in certain populations in which seasonal rhythmic changes of frequency of the arrangements occur with greater or less regularity. Evidence obtained by systematic collections for five years at two stations in the San Jacinto Mountains shows a change of structural heterozygosity in both the third and X chromosomes that is correlated with rhythmic changes in frequency of the arrangements present. Experimental evidence from other species suggests that the consequent decrease of crossing over in these chromosomes would be accompanied by an increase of crossing over in the homozygous second and fourth chromosomes, that it may increase differentially in some of their parts, and that this increase is accompanied by an increase of variability. Assuming that the whole genome is an operative unit in respect of crossing over, the hypothesis is presented that the rhythmic changes of the different arrangements are induced by an adaptive advantage of certain crossover products in the second and fourth chromosomes combined with those in certain arrangements in the third and X chromosomes. This advantage is related to seasonal environmental differences at the two stations. Thus, the inversion system is regarded as a mechanism that preserves the adaptability of a species by mediating recombination to the exigencies of different and changing environments.

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CHANGES IN AN INVERSION SYSTEM DURING A HUNDRED GENERATIONS [1]

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INTRODUCTION

An almost continuous record has been made since 1939 of the frequencies of different gene arrangements in populations of *Drosophila pseudoobscura* in the San Jacinto Mountains near Los Angeles (Dobzhansky, 1943, 1947; Epling, Mitchell and Mattoni, 1953, 1957). If we assume that a generation of this species has an average life span of about a month, this period of observation covers not less than 100 generations. More than 40,000 third chromosomes have been examined and a comparable number of X chromosomes, these being the two chromosomes in which inversions occur in detectable frequencies. The samples have been made at monthly intervals as far as possible and the record reflects the changes that have occurred each year for a number of years.

The basic analysis of each gene arrangement has been published by Dobzhansky (Dobzhansky and Epling, 1944). Three other arrangements are being described in this paper. They bring the number known in the third chromosome to twenty, of which twelve occur in the populations referred to. Two arrangements are known in the X chromosome. One of these, known as Sex Ratio, is associated with a genetic factor that causes an alteration of the sex ratio and produces mostly females in matings of males that carry it.

The frequencies of the different arrangements vary greatly. The Standard arrangement of the X chromosome (STX) is found in roughly 85–90% of most samples, the sex ratio arrangement (SR) making up the difference. The Standard arrangement of the third chromosome (ST) is found in 35–45%, on the average. The other principal arrangements are known as Arrowhead (AR), Chiricahua (CH), Pikes Peak (PP) and Treeline (TL). They range in mean frequency from about 25–30% to 4 or 5%. The data of frequencies for the whole period are summarized in Table 1 of Epling, Mitchell and Mattoni (1953) and in Tables 1–4 of Epling, Mitchell and Mattoni (1957). The names of other less frequent arrangements will be found in figure 4 of this paper and their frequencies in the area are shown in figure 3 of this paper.

LONG TERM FLUCTUATIONS

A comparison has been made below of the mean frequencies of the principal arrangements at Pinon Flat and Keen Camp from 1939–1942 with those from 1952–1956. These are periods when collections were most regularly made and

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	ST		AR		CH		PP	TL
	1939–1942	1952–1956	1939–1942	1952–1956	1939–1942	1952–1956	1952–1956	1952–1956
Pinon Flat	.416	.477	.249	.215	.280	.154	.087	.053
Vandeventer Flat	—	.451	—	.230	—	.175	.088	.055
Keen Camp	.325	.360	.234	.256	.394	.282	.066	.036

when samples were the largest. The comparison indicates that the mean relative frequency of each arrangement has not changed greatly during the whole period of collection. Comparison of the whole array of samples shows, however, that fluctuations in relative frequency of three kinds are found over and above those expected of sampling error: irregular monthly and yearly fluctuations, predictable seasonal fluctuations, and regular but unpredictable fluctuations of longer or shorter term that may last for several years. The seasonal fluctuations, described first by Dobzhansky (1943), are rhythmic and recur each year with remarkable regularity in some populations such as that at Pinon Flat, but with less regularity at Vandeventer Flat and not at all at Keen Camp. They are discussed in view of recent data by Epling, Mitchell and Mattoni (1957). The environmental characteristics of the collecting stations to which we shall refer are also described there. Our purpose in the present paper is to discuss the long term fluctuations in these populations, of which one segment from 1939 to 1946 has been described by Dobzhansky (1947).

We refer now to figure 1. The curves in this figure show the mean trends of frequency since 1939. They were obtained as follows. The mean frequency of each arrangement in all samples since 1939 was computed from the basic information in the tables of data referred to. Using these means as 100, the per cent to which each yearly mean departed from its long term mean was computed and graphed. The curves shown in figure 1 are the smoothed curves taken from these graphs. Inspecting them, we can compare the mean trends of frequency of each arrangement and ascertain whether the trend of one is necessarily dependent on that of another. We can also compare the trends at one station with those at another. The trends for SR are shown only for 1939–1942 and from 1952–1956. The few samples made in 1945 and 1946 are not representative of the whole collecting season and comparisons would be misleading because of the presence of a seasonal rhythm in the arrangements of both chromosomes at Pinon Flat. The frequencies of SR were not obtained by us from 1948–1951.

If we take first the trends of ST and AR shown in figure 1, we find that they have been generally inverse to each other at Pinon Flat, and that their general relations have been much the same at both stations. If we compare the trends of CH at the two stations, we find that they, also, are similar, but with a greater fluctuation at Piñon Flat. They can also be said to be inverse to the trend of ST, but the dependence is not very close. The trends of PP have been similar at both stations. Its frequency prior to 1951 has not been recorded because it was first detected in the San Jacinto Mountains that year. If it existed in this area during the earlier years, then its frequency must have been so low as to make it undetectable. Its trend has been inverse to that of CH. Comparing now, the trends of SR, we find that it also has been similar at both stations. The scanty data for 1945 and 1946 (which are not graphed) indicate that it may have increased during this middle period. It does not appear to have been closely dependent on any of the arrangements of the third chromosome in the earlier period, unless it was inversely proportional to CH, with which it has been more or less directly proportional from 1952–1956. Its decrease from 1939 to 1942 at Keen Camp may have been dependent on the decrease of AR. If so, the situation was the reverse at Pinon Flat.

These comparisons suggest that each arrangement has fluctuated independently of any other during this period in this area. This biological independence of change, over and above the requirements of mathematical dependence induced by the means of comparison, is strengthened by observations made by Dobzhansky (1952, 1956) at Mather in the central Sierra Nevada and by observations made

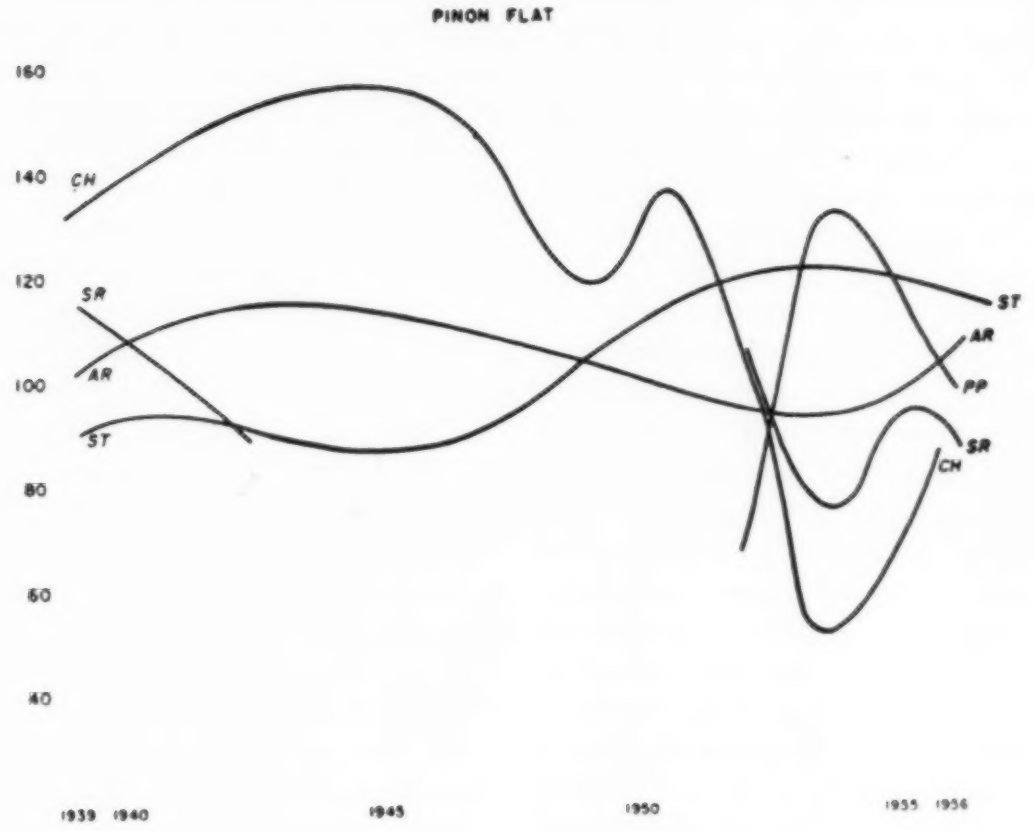

FIG. 1. Trends of relative frequency of the four most abundant arrangements of the third chromosome and the SR arrangement of the X chromosome at Pinon Flat and Keen Camp, based on the mean frequency of each since 1939, indicated by 100. See text for explanation.

in the Charleston Mountains near Las Vegas, Nevada. Pikes Peak has been known in the central Sierra Nevada of California since 1941 (Dobzhansky and Epling, 1944). In recent years it has increased in frequency inversely in relation to a decrease of CH (Dobzhansky, 1956) just as in southern California from 1952–1956. The only collections made in the Charleston Mountains have been made by Dobzhansky in 1937 (Dobzhansky and Epling, 1944) and by Epling and Mattoni in 1955. No PP was recorded there in the earlier collections but in 1955, it had a frequency of .063. CH had dropped from .191 to .094. The frequencies reported in both years are as follows:

	N	ST	AR	CH	PP	TL
1937	256	.121	.688	.191	—	—
1955	126	.244	.559	.094	.063	.039

The collections were made in both instances in the same month of the year (April) and at the same place. Now, while PP and CH were changing inversely, ST and AR were maintaining much the same relations in southern California. But in the Charleston Mountains when PP and CH were changing inversely with much the same frequencies, ST doubled in frequency and AR dropped. A similar inverse relation between PP and CH at Mather from 1945–1954 has also been accompanied by a shift between ST and AR but in this case, AR increased relative to ST (Dobzhansky, 1956).

These data indicate that although the over-all pattern of relative frequencies of the different arrangements appears to have a considerable permanance in each locality, its components are continually fluctuating with more or less regularity, but not in a predictable way. The fluctuations are also accompanied by changes in structural heterozygosity in the third and X chromosomes and the multiple heterozygotes of both. This is shown in table 1. The raw data on which this table is based will be found in Table 4 of Epling, Mitchell and Mattoni (1957). The estimates arrived at were obtained in the same way as those of Table 5 of that publication. The frequencies of heterozygotes in the third chromosome have not varied significantly during the fluctuation; heterozygosity in the X chromosome on the other hand, decreased regularly until 1954 and has increased since. The over-all structural heterozygosity expected in both chromosomes, which would include multiple heterozygotes, was significantly lower in 1953 and 1954 than in the other years, and free recombination would have increased accordingly. Epling, Mitchell and Mattoni (1957) have shown that an over-all increase has occurred regularly during this period during the seasonal rhythm at Pinon Flat, resulting from an increase of structural heterozygosity in both chromosomes. At an adjacent station, Vandeventer Flat, a similar increase resulted chiefly from an

TABLE 1. *Mean yearly frequencies of inversion heterozygotes at Keen Camp during 1952–1956 estimated for females*

	1952	1953	1954	1955	1956
Expected frequencies of third chromosome heterozygotes					
A	.716	.718	.724	.699	.719
Expected frequencies of X chromosome heterozygotes					
B	.252	.194	.175	.281	.191
Expected frequencies of females heterozygous for both chromosomes					
C	.180	.129	.127	.196	.137
Expected frequencies of females heterozygous only for the third chromosome					
D	.536	.579	.597	.503	.582
Expected frequencies of females heterozygous only for the X chromosome					
E	.072	.055	.048	.085	.054
Estimated over-all heterozygosity of females					
F	.788	.773	.772	.784	.783

increase of heterozygosity in the X chromosome. The combined data suggest an association of some sort of heterozygosity in the two chromosomes, even though they also act independently, and ascribe it to the relation of heterozygosity to crossing over and recombination in the whole genome.

Increase of the Number of Arrangements

Until 1951 the inversion system in the third chromosome in the San Jacinto Mountains apparently consisted of seven arrangements: those we have already referred to, and two others of very low frequency, Santa Cruz and San Jacinto (fig. 2). Santa Cruz has maintained a frequency of 0.3–0.4% for the whole period. A third arrangement named Vandeventer in this paper was also apparently present. It had been seen once before (in 1942), but the observation was not published (Dobzhansky, orally). In 1951, three Pikes Peak arrangements were found in a July sample at Pinon Flat after at least 22,000 chromosomes from this station had been examined. This arrangement was new to the area, and indeed, to southern California. By March of 1952, it increased to 10.8% and its mean frequency for the whole year at both stations was 6.0% (fig. 2). Thus, it increased from negligible proportions prior to 1951 to the fourth place in frequency in 1952, within a few generations. Pikes Peak is a readily detectable arrangement and if it existed in the area prior to its discovery, it must have had a frequency comparable to that of San Jacinto. In 1953 four other arrangements were found that had not previously been observed, Estes Park and Olympic, which are widespread elsewhere, and two others, previously undescribed, named Vandeventer and Thomas in this paper. Vandeventer, as noted, had been seen by Dobzhansky in 1942. In 1954, another new arrangement, Pinon, was found, and San Jacinto was found the second time in 1956. The frequencies of all these are shown in figure 2, and the new arrangements are illustrated in figure 3.

To summarize, the number of arrangements in this area, approximately doubled in a few generations. Some, like Pikes Peak and Olympic, increased significantly in frequency for a while. The latter appears to have decreased in 1955 and 1956 to the extent to suggest that it may presently "disappear." Pikes Peak, however, which also appeared suddenly seems to

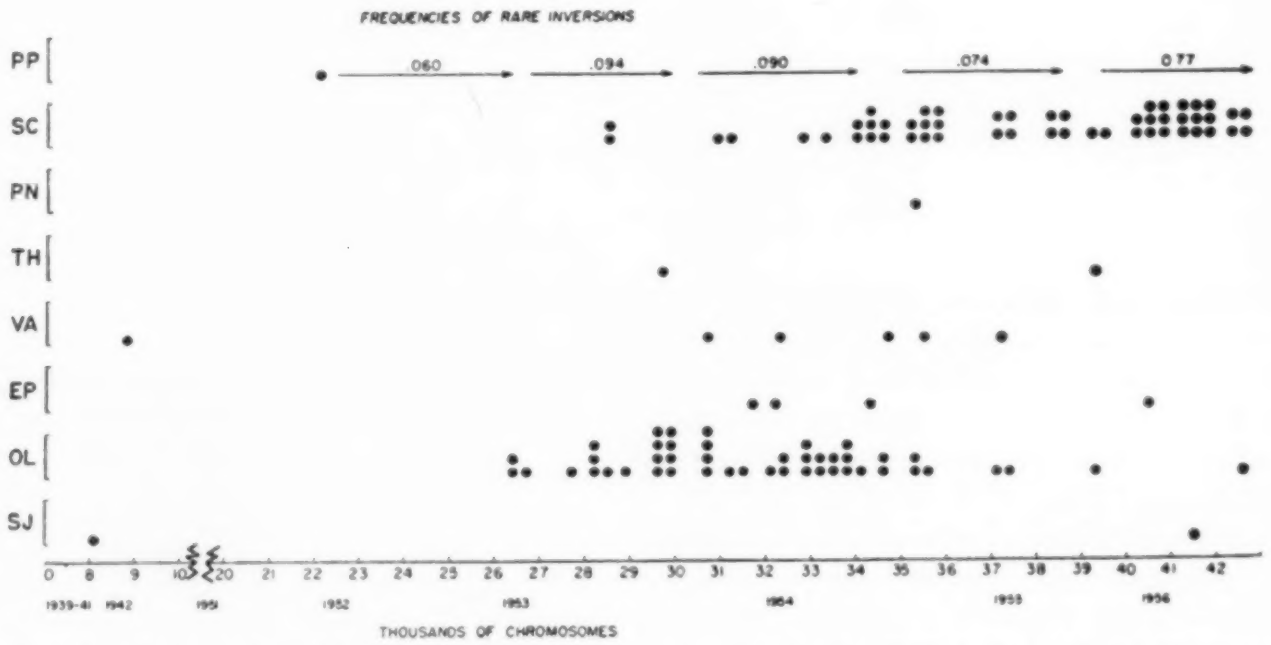

Fig. 2. Frequencies of less abundant gene arrangements in the San Jacinto Mountains since 1939. Santa Cruz and Treeline are shown only since 1952 for comparison. Pikes Peak is indicated by percentages. Discussion in the text.

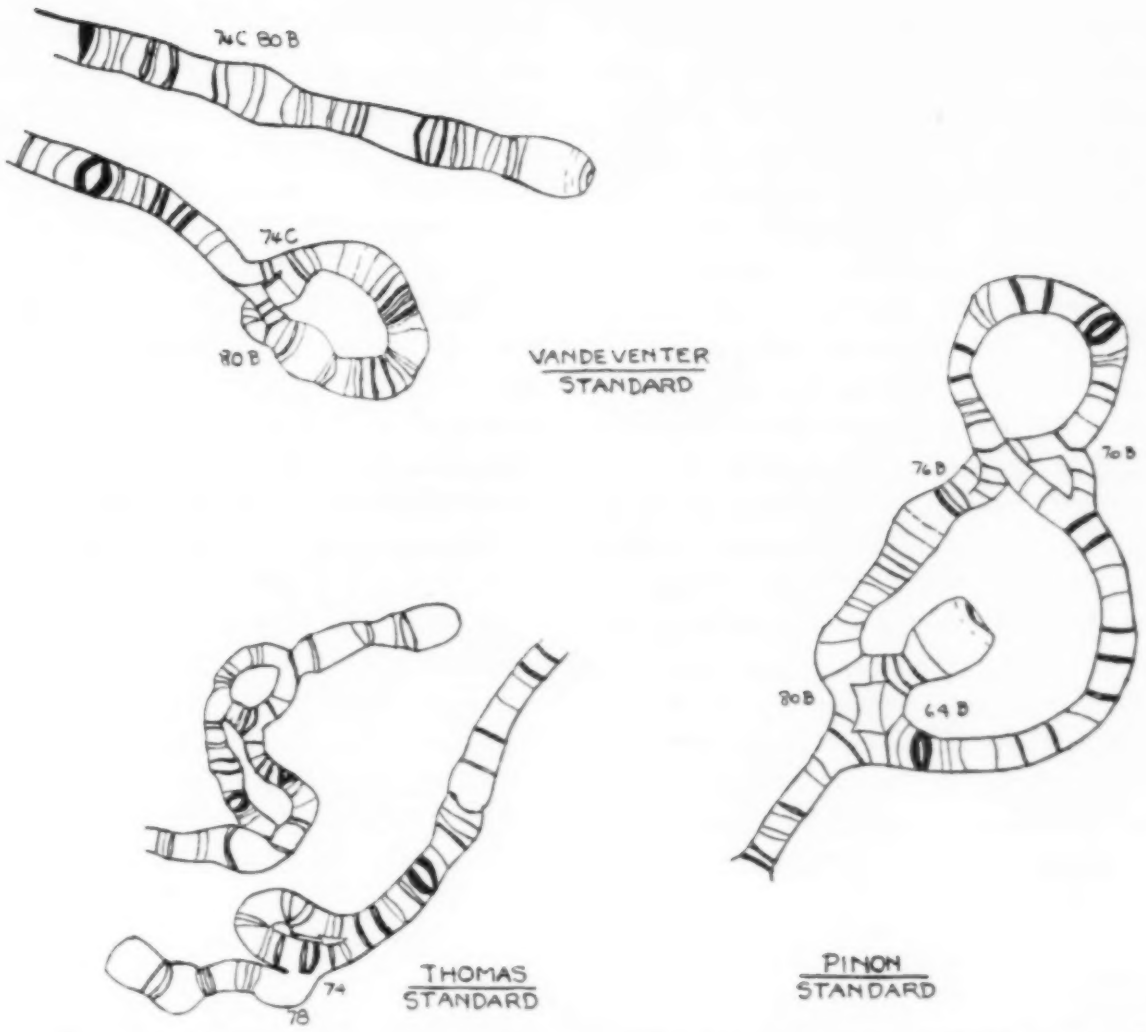

FIG. 3. Illustrations of heterozygotes of three undescribed arrangements, Vandeventer, Pinon and Thomas, with Standard, and the distal part of the homozygotes of Vandeventer.

be maintaining itself at the level it soon established.

Several explanations are possible. First, Olympic, Pikes Peak and Estes Park may have been introduced into the area either by migration or by long distance dispersal. The first two arrangements are known further north in California, and Pikes Peak is known to have increased in frequency in the central Sierra Nevada since 1941 (Dobzhansky in Dobzhansky and Epling, 1944; Dobzhansky, 1956). Furthermore, Estes Park has been found recently in northern Nevada near Elko (Dobzhansky, orally) and Pikes Peak has been found in the Charleston Mountains near Las Vegas, where it was not known before. They might have spread recently. Or, what seems to us to be the more remote possibility, all three might have been introduced simultaneously by automobile traffic along the main highway from Texas. Second, these arrangements may have been present during the whole period and have only recently risen to detectable proportions, as a result of long term fluctuations similar to those of arrangements present in greater frequency. Third, the possibility also exists that some have arisen by independent mutation. There are no data that permit a decision between these alternatives except those that suggest the possibility of mutation—but this need not be applicable to the present situation.

The positions in the third chromosome at which breakages have been recorded shows that they are not randomly distributed. More breaks occur in the distal than in the proximal half. Furthermore, what seems to be the same break, as far as cytological examination can assure us, has occurred independently on more than

one occasion (Figs. 4 and 5). Figure 4 has been constructed as follows. The arrangements known in both *D. pseudoobscura* and its sibling species *D. persimilis* are listed along the ordinate. One will recall that both species have the Standard arrangement in common. The series follows the phylogeny shown in figure 5 which has been adapted from Dobzhansky (in Dobzhansky and Epling, 1944). The various numbered sections and subsections of the Standard chromosome appear along the abscissa of figure 4 (Dobzhansky and Sturtevant, 1938). The subsection at which each break has been recorded is indicated by an oval opposite the arrangement name which is connected with the section by a vertical line. The distribution of breaks over the whole chromosome has been summarized by the bar diagrams in the lower right hand corner. In the first case, the 68 lettered sections were divided evenly into ten groups, and in the second, into five. The distribution of the breaks in each group are indicated by the bars. The breaks clearly occur more often in the distal half, which corresponds to Helfer's evidence from irradiated stocks (1941). A similar pattern of distributions will also be found in the Hypothetical and Santa Cruz arrangements if they are analyzed in the same way. This is mentioned because any one of the three may have been the initial arrangement in the phylogeny (fig. 4; Dobzhansky in Dobzhansky and Epling, 1944).

Comparison of figures 4 and 5 and the distribution maps published in Dobzhansky and Epling (1944) will show, first, that the same point of breakage occurs in arrangements found in widely separated and climatically different parts of the species area. That in 64C, for example,

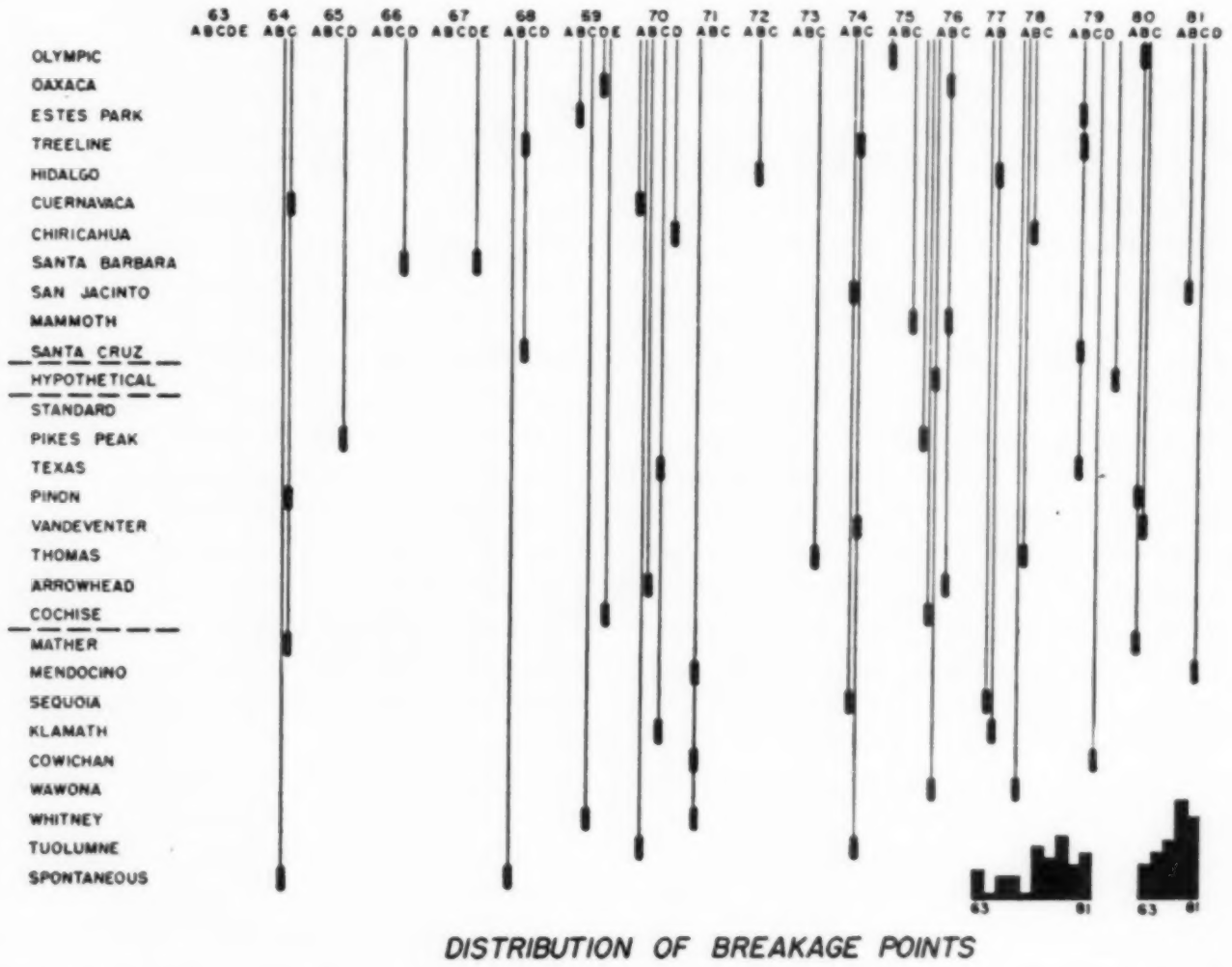

FIG. 4. A diagram to illustrate the relation of the breakage points of the different arrangements of the third chromosome of *D. pseudoobscura* and its sibling species, *D. persimilis*. Description in the text.

FIG. 5. Phylogeny of the arrangements of *D. pseudoobscura* and *D. persimilis*.

occurs in the Cuernavaca arrangement found in central Mexico and Guatemala, and in Pinon, and Mather, found in California. That in 70C occurs in Klamath, which ranges from British Columbia to central California, and in Texas, found in a single individual in that state. That in 76B occurs in Arrowhead, which ranges from British Columbia to northern Mexico, and in Oaxaca, which has much the same range as Cuernavaca. Second, the same point of breakage occurs in arrangements of different phylogeny and would have been formed independently after a series of steps from their antecedents. The same examples illustrate this point. The break in 64C occurs in a derivative of Santa Cruz (Cuernavaca), in a derivative of Arrowhead (Pinon), and in a derivative of Standard of *D. persimilis* (Mather). Cuernavaca is separated in the phylogeny by five steps from Mather and six from Pinon. That in 70C occurs in a derivative of Standard of *D. persimilis* (Klamath) and in a derivative of Pikes Peak (Texas). Klamath and Texas are separated by three steps. That in 76B occurs in a derivative of Standard (Arrowhead) and in a derivative of Treeline (Oaxaca). Arrowhead and Oaxaca are separated by five steps. The break between 74B and C occurs in a derivative of Standard (Vandeventer), a derivative of Whitney (Tuoloumne) and a derivative of Santa Cruz (Treeline). Treeline and Tuolumne are separated by six steps. Thus, it would appear that breaks in each series have occurred independently and possibly in different parts of the species area. Whether the breakage points that correspond are in fact identical is uncertain. They appear to be in many cases. In others they are clearly different but very close. In any case, two arrangements with substantially the same gene order would be formed should two breaks occur simultaneously at the same or similar points of a given arrangement. We know of one instance in nature that suggests this possibility. The breakage points of Mather and Pinon are at least very similar. The first was formed directly by a single inversion in the Standard arrangement of *D. persimilis*

(Dobzhansky, 1948), the latter was formed directly by a single inversion in the Arrowhead arrangement of *D. pseudoobscura,* as shown by the included Arrowhead inversion (fig. 5). Had the Pinon breaks occurred in Standard instead of Arrowhead, substantially the same arrangement would have been formed independently in two widely separated areas.

We have been unable to directly compare Pinon and Mather inasmuch as a stock of Mather was not preserved. We have, however, compared cytologically eight strains of Pikes Peak from the San Jacinto Mountains with five from Texas, kindly sent by Professor Wilson Stone, and have detected no cytological differences in twenty strains of the crossed homozygotes.

Conclusion and Summary

The system of frequencies of the gene arrangements that are present in the populations of *Drosophila pseudoobscura* in the San Jacinto Mountains appears to have had a certain stability during 100 generations. Each of the principal arrangements has fluctuated from its mean during this period in a fairly regular way. The trends of any given arrangement do not appear to have been dependent on another for the whole period, although one may be more or less directly or inversely proportionate to another for a part of it. A fluctuation in structural heterozygosity has been observed since 1952.

Since 1951, the number of arrangements known has increased from seven to twelve. One of those newly detected increased during a few generations from negligible proportions to roughly 6%, becoming one of the more common arrangements. The others have persisted in low frequency and are apparently decreasing to the point of disappearance. Their detection and increase can be accounted for on the ground of long term fluctuations similar to those of the long established arrangements. The possibility exists, however, that some may have arisen independently by mutation. Evidence is presented to show that the breaks in the third chromosome from which the known inversions have been formed tend to be localized and not at random, that they have recurred independently during the phylogeny of the inversion system, and that a similar pair of simultaneous breaks has occurred independently in different arrangements.

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NOTES AND COMMENTS

THE USEFULNESS OF SCHOLANDER'S VIEWS ON ADAPTIVE INSULATION OF ANIMALS

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Scholander's (1955) exposition of the usefulness of adaptive insulation in considering the evolution of homoiothermous animals necessarily included denial of significance to the climatic rules attributed to Bergmann (1847) and Allen (1906). Vigorous replies from Newman (1956) and Mayr (1956) insist that the climatic rules can be useful along with Scholander's physiological observations.

I hope that the clarifying influence of this enlightening controversy among my admired colleagues will be sustained by my comments. It is with pleasurable anticipation of the value of debate that I enter reasons which deny significance to some observations by which Newman (1956) and Mayr (1956) would support the use of the climatic rules as indicators of the progress of evolution in homoiothermous animals. I agree with Scholander that there is no reason for sustaining the climatic rules. On the positive side of the argument I will comment upon some recently developed information about adaptation to climate which has been prepared by the use of the propositions which Scholander enunciates.

Scholander has pointed out essential errors in the thermal bases upon which Bergmann's and Allen's rules rest. I am in a position to point out a fault of evidence in a widely used report which states that climate has had an adaptive influence upon the morphology of Eskimos. I refer to Newman's (1956) quotation of Coon, Garn and Birdsell (1950) as to the commonness of amputations of Eskimos' fingers from freezing. During ten years I have often worked with Alaskan Eskimos without noticing that amputations for cold were common. Stimulated by Coon's statement, Dr. A. B. Colyar, Dr. E. M. Scott [1] and I discussed the importance of obtaining statistics upon the extent and effects of cold injury among arctic people. Dr. Colyar has kindly informed me of the indications from his preliminary inquiries. Replies from physicians in charge of hospitals in northern Alaska indicate that 2 or 3 admissions for frostbite occur annually from each of several population groups of about 1000 Eskimos. I have seen no report upon amputations but suspect them to be fewer, since several of the admissions were reported discharged as recovered.

For some years now significant injuries in these areas have reached hospital attention. Reporting physicians mention frostbite as associated with accidents to aircraft and immersion so that to determine the natural incidence of frostbite requires further scrutiny. The treatment of cold injury is an important consideration for physicians in regard for the health of individuals. But frostbite does not appear to be more than a minor concern for public health among arctic Alaskan residents. Since its incidence can be defined it will probably be determined as a useful statistic in planning for health. But there is not the right kind of evidence concerning cold injury to suggest probability that the evolution of morphology in the extremities of Eskimos has been adaptively molded by cold.

Scholander has stated that Eskimos are adapted to cold in a large measure by their clothing and housing. There is some adjustment by individual and social behavior, and but a small degree of adaptation remains to be sought in their morphology and reactions. The elucidation of the small human adjustments to cold is a very important physiological task. Whether these adjustments are found to be adaptations according to the evolutionist's usage will be of critical importance in their physiological definition, in the consideration of health and in views upon the origin of racial distinctions. It is difficult, however, to observe whether Eskimos differ from white men in their reactions to cold because experienced arctic residents so carefully shield their physiological adjustment to cold within the small human range of tolerance.

Newman's (1956) reference to variations reported in basal metabolism relate them in some cases to climate or other circumstance and in some cases to race. The small differences between Eskimos and white men are contradicted in other reports. Rodahl's (1952) survey showed that results were easily affected by the procedures used in metabolic measurement of Eskimos. Erickson's (1949) careful measurements of respiratory and metabolic characteristics of young Eskimos and white men at Point Barrow did not show racial distinctions. Scho-

[1] Medical Officer in Charge and Biochemist, respectively, Arctic Health Research Center.

lander and Krog (1956) did not find the metabolism of Lapps, an arctic people, distinctive. In looking at many other reports I have felt that Du Bois' (1936) critical view of the variations reported in the metabolism of different races and in various circumstances must now be applied even more critically, for while observations have multiplied, the standards of procedure and even of accurate performance have also changed. Du Bois did not think that the small variations reported in basal metabolism showed superiority. Without demonstrable advantage a variation cannot be presumed adaptive to climate.

The climatically adaptive value of even considerable variation in metabolic rate would be small. If the metabolism of man is doubled he had a critical temperature when naked about 17° C. instead of the usual 27° C. (Erikson, Krog, Anderson and Scholander, 1956). If it were to be established that the basal metabolism of Eskimos exceeds that of white men by 25%, the greater heat production would give to the Eskimo a critical temperature 2.5° C. lower, or about 24.0° C. That is a small step toward adaptation to arctic winter air at −40° C., amounting to 4% of the distance toward the protection afforded by good fur clothing. It would not be accomplished without extra cost for food and would involve a large change in the speed of the metabolic machinery. To postulate adaptive advantage in this minor protection from cold is not reasonable without considering its size and the consequent results from derangement of a metabolic economy which is so remarkable for its common stability. The small advantage of a constant high basal metabolism in winter would be a relatively larger disadvantage in warm summer weather and during exercise.

I cannot agree with Mayr (1956) that adaptive significance can be shown in the small natural variation in the dimensions of animals by use of the climatic rules. With change in geometric dimensions many other energetic relations beside heat are affected. As Scholander (1956) so aptly illustrated, large size would limit the free movement of a large aquatic mammal through its aquatic habitat when it becomes about 90% ice in the northern winter. Disposable muscular power increases with mass and the velocity of extremities is much affected by their length. Perhaps it is the balance of these influences which keeps animals so generally within narrow limits of variation in size. It is certain, however, that many beside thermal relations are affected by size.

Two or even ten fold variations in mass are visible among average adult dimensions of mammals of one sex of a species. Birds of a species seem to show small variation in size, perhaps because of the exacting dimensional limitations of aerodynamics. Such extremities as ears, noses and tails, which I will lightly term decorative appendages to avoid considering their specialized functions, may differ several times in area within a species. Limbs are limited in variability. If the dimensional variations seen were combined for the conservation of heat, they could not improve it by a few per cent. As Scholander shows, geographical variations in climate require and consistently find among animals 10 fold changes which is provided by insulation for the conservation of heat.

Favoring relations between surface and mass might in an otherwise unaffected environmental relation extend the natural range of some forms over a temperature range of a degree in winter. Within the narrow limit of this temperature the population's range could be favored for extending northward. But in cold regions there is also a warm season during which the heat conserving effect of a surface to mass relation would correspondingly oppose the dissipation of heat. Exercise is also limited by the rate at which heat can be dissipated. The difficulties encountered in postulating adaptive value for the inflexible surface-mass relations contributed importantly to development of the idea that flexible insulation best described conditions of thermal exchange between the homoiothermous animal and its surroundings (Scholander, Hock, Walters, Johnson and Irving, 1950).

Analysis of the components of the overall insulation of animals is defining and locating them in anatomical and physiological terms so that we can begin to add thermal magnitudes to the suspected value of these adaptations. The thickness of fur largely explains (and that is rarely a proper term even among physiologists) how the well adapted terrestrial mammals live in the arctic and in the tropics with the same metabolic cost for heating (Scholander, Hock, Walters and Irving, 1950). But furless swine live in freezing air in Alaska with no greater metabolic expense for heat than is required of swine in a warm climate (Irving, Peyton and Monson, 1956). Hair seals, which are uninsulated from water by their wettable hair, are nevertheless so well insulated that even in icy water they scarcely use extra metabolism for heat (Irving and Hart, 1956). Because ice water has about ten times the cooling power of the coldest naturally occurring air, it is now safe to consider it general that the metabolism of adapted mammals is protected by their various insulating systems to operate at specific metabolic levels in the different climates which they inhabit.

The chief insulator of the bare skinned mammals was foreshadowed in the observed coldness of the thinly insulated extremities of arctic

mammals (Irving and Krog, 1955). This coldness extends in swine and seals over the entire body and involves a considerable mass of underlying tissues in a gradient of temperature which depicts their insulative action (Irving, 1956; Hart and Irving, 1956). Scholander (1955) has described vascular heat exchangers as probably the important physiological devices employed in the variable insulation of bare extremities and has demonstrated their pattern in porpoises (Scholander and Schevill, 1955) and in sloths (Scholander and Krog, 1956).

The cooling of superficial tissues in birds and mammals for the conservation of heat shows that homoiothermism is sustained by the capability of some tissues for heterothermism (Irving, 1956). For heterothermous tissues to operate in association with a homoiothermous body requires special thermal characteristics such as have been shown in the specialized ability of the distal parts of nerves from the cool legs of gulls to function while cold (Chatfield, Lyman and Irving, 1953) and in the low melting points of the fats of the extremities of mammals (Irving, Schmidt-Nielsen and Abrahamsen, 1956). The visible integration of heterothermous tissues in an individual with a warm homoiothermous center offers thermal considerations as a new aid in viewing how integration of life is effected within an individual animal. Some essential characters suitable for view as to their heredity and evolution can be revealed by their experimental definition in the well regulated thermal differences of the heterothermous tissues concerned with insulation.

We hoped that contradiction of the significance attributed to Bergmann's rule would be acceptable if it came from physiologists, who deserve a share of blame for the exaggerated attention given to minor thermal factors in biology and anthropology. It will follow only after longer controversy that the "law of surfaces" will be removed as an even heavier incubus upon physiology. Although the elimination of error is a bitter process, in freedom from the erroneous climatic rules we can already perceive useful new aplications of physiological method to the greatest science, biology, in which evolution requires man's noblest effort at thinking.

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AN EXPERIMENTAL INVESTIGATION OF JUDGMENTS CONCERNING GENERA AND SPECIES

EDGAR ANDERSON [1]

We may consider species and genera in two quite different ways: either as groups of individuals (which may or may not exist in nature) or as concepts concerning these groupings which exist in the minds of systematists. Discussions concerning the species problem are frequently at cross purposes because these distinctions have not been clarified before proceeding further. Though species in the latter sense are, to use Asa Gray's phrase, nothing but judgments, we may study them in a truly experimental way by performing objective experiments in the domain of applied psychology. A formal report of one such experiment is given below and is discussed in the light of several less formal attacks on the same problem.

MATERIALS AND METHODS

From preliminary experiments in which unlabelled specimens were given to foreign graduate students in taxonomy not yet familiar with the American flora, it seemed that on a non-articulate level there must be some agreement about species and genera. It was discovered by trial, that if one picked out "good" genera and "good species foreign experts with no knowledge of our flora (experts furthermore with no specialized training with that particular family of flowering plants) would usually divide the unlabelled specimens into something close to the arrangement of genera and species which had previously been worked out by American taxonomists. Through the kind offices of Dr. Robert Cooper a package of herbarium specimens was forwarded to three New Zealand systematists for judgments as to the numbers and kinds of taxa which it included. The central core of the instructions accompanying the sheets was as follows: "What I should like is your judgment after examining the specimens as to how many species, varieties, and genera are involved and (by the numbers) how these 16 plants are to be classified. I am not at all interested in having you identify them as to family or genus. Quite the opposite; I hope you will not refer to the literature until after you have dealt with the specimens (if then). This much I can assure you. They were chosen because their classification presents no problems and has never been under dispute. If, for instance, they belong to five species in two genera, then the species are all clear cut and the genera are universally recognized as coherent, distinct genera. If there are any varieties or sub-species present in the material then they too are well-marked variants but not so distinct as to merit specific recognition in the opinion of anyone who has dealt with them."

It should be noted that the recipients were given no clue as to the number of genera, species, and varieties included in the sample; they were if anything encouraged to believe that more than one genus was involved. They had no information about geographical distribution other than that the specimens all came from North America. They were however assured that no problem genera, nor hybrid swarms, nor apomictic groups of doubtful relationship, had been included in the sample.

RESULTS

Three different systematists examined the sheets in New Zealand and reported their results. One was a distinguished monographer of another family of plants. His judgments were arrived at completely independently of the other two. They however discussed the specimens with each other though they arrived at their final judgments independently. One of these men had done extensive field and experimental work with speciation in a difficult group of flowering plants, the third was an algologist. It is an interesting point that his judgments most nearly agree with the dispositions made of these plants by all the American taxonomists who have worked with them in the field and the herbarium. The disposition of the 16 specimens by the three experts is presented in tabular form in table 1.

The specimens selected for the experiment were two species of Uvularia from eastern North America, *U. grandiflora* and *U. perfoliata*, common liliaceous perennials in our spring woodland flora. The specimens were discards from a big herbarium and were reasonably complete but without any notes as to locality, date, color, etc. Some of each species were in flower and some in fruit but there were no complete specimens showing the same plant in both these aspects.

It will be noted from Table I that, though all three men had been led to expect samples of more than one genus, the specimens were all correctly determined as belonging to the same genus. This same result had been ob-

[1] The above paper was written during my tenure of a Fellowship from the Guggenheim Foundation and while I was a visiting Fellow of Princeton University.

TABLE 1

The first letter indicates the genus, the second the species, the number indicates varieties where recognized. X–B–1 and X–B–2, for instance, indicate two different varieties, 1 and 2, of species B, belonging to genus X. It will be noted that all three collaborators judged all the specimens to belong to one genus.

Plant number	Monographer	Experimental taxonomist	Algologist
	U. grandiflora		
3	X–C	X–E	X–E
5	X–B	X–E	X–E
6	X–E	X–B–1	X–B–1
7	X–D	X–B–2	X–B
9	X–E	X–B–1	X–B–1
10	X–D	X–B–2	X–B
11	X–E	X–B–1	X–B–1
13	X–D	X–B–2	X–B
15	X–D	X–E	X–E
16	X–4	X–B–2	X–B
	U. perfoliata		
1	X–D	X–B–3	X–A
2	X–D	X–B–3	X–A
4	X–D	X–C	X–C
8	X–A	X–C	X–C
12	X–A	X–C	X–C
14	X–D	X–B–3	X–A

tained in all the preliminary experiments. When Uvularia specimens had been used, if the foreign expert were specifically asked "Are you sure these belong to one genus?," he usually replied with some such remark as: "why, of course they do." Note furthermore that in general the division into species follows the conclusions of American botanists, though all three New Zealanders attempted to divide the sample into more species than can be recognized with larger and better collections and after extensive field experience. This is partly because the small sample included a rather high proportion of collections from the regions where the two species overlap and where Dietz (1953) has shown that there is slight (but widespread) introgression between the two species, particularly in those sites where they grow close to one another.

From these and similar experiments one is forced to conclude that there is a phenomenon here which, since it relates to a basic biological problem, is worthy of prolonged and careful study. It would require consultation and further research merely to set up the kinds of tests which could be meaningful both to psychologists and biologists. These preliminary results do suggest, however, that for stable floras, such as those of eastern North America, there is some kind of general agreement (apparently inarticulate and intuitive) among the systematists of the world as to the nature of species differences and generic differences. Another set of data supporting the same conclusion can be drawn by correlating judgments as to species and genera recognized in the same flora by rival groups of systematists. For this purpose Fernald's last revision (1950) of Gray's Manual and Gleason's (1952) revision of Britton and Brown are excellent experimental material. Though Gleason and Fernald met each other on terms of mutual respect they worked at taxonomic centers between which there had been keen professional rivalry and conspicuous, intense, and prolonged difference of opinion about nomenclatorial procedure, the generic concept, and other matters. There were other differences in the backgrounds of the two men. Fernald had since his undergraduate student days been closely associated with taxonomy. Though he worked much in the field his companions were nearly all professional or amateur systematists. Gleason was originally a professional ecologist; he continued these interests and associations even after he became immersed in the taxonomic problems of the plants he had previously studied ecologically. Yet if we open these two floras side by side we find it hard to pick a family or a genus in which they do not agree (extensive spot checks have been made) at least 8 or 9 times out of 10 when they cover the same area.

This is of course a rather limiting case. The flora of eastern North America is relatively stable and it has been well and intensively studied for over a century. In California speciation is much more intricate for all kinds of organisms. Microclimates are much more highly developed. Tertiary and Pleistocene sea-level, rainfall pattern, and mountain height changes have been extreme and have led to complicated speciation patterns. The flora furthermore has not been so intensively studied for so long a time, yet when one takes two of the standard floras of the state and deals with comparable areas he finds complete agreement 4 to 6 times out of 10 and differences only in detail 2 to 3 times out of 10. Acrimonius disputes between dedicated naturalists may have obscured a significant fact. The judgment of taxonomists must somehow be more objective than they themselves sometimes lead us to believe. The disputes were over the small percentage of instances in which they did not agree. Taxonomic intuition, though it may seem mystically non-scientific to those without much of it, must be based upon some kind of sense impressions. By observation and experiment (including experiments upon taxonomists as well as upon the organisms they work with) it should be possible to dig into this basic scientific problem.

By making objective comprehensive surveys of the morphological nature of species differences (Anderson and Whitaker, 1934) and by making objective tests, such as those reported here, on the reactions of systematists to such differences, we should be able to get down to a new level of understanding of species-genus-family relationships. To me it seems quite likely that there may be basic generalizations concerning species and genera which may be reached by some such route. Certainly the known physico-chemical complexities of the germplasm must so canalize its rates and kinds of change that the products of the world's germplasms would have to be severely canalized by these forces alone, even were there not (as we know there are) further canalizing forces outside the germplasm.

From morphological study alone one can turn up vague indications of such generalizations. Since 1924, by various experimental techniques I have been attempting to study the nature of species differences. Though, as Linnaeus said, the differences are where you find them, they are certainly not distributed at random (Anderson and Whitaker, 1934). During the last two decades my students and I have made objective surveys of variation in various species and species groups. We have attempted to survey the totality of the difference between species in so far as it was humanly possible and to measure and record all the elementary differences of which it was composed. For the best of these studies the major portion of the morphological differences between the species under observation must have been brought into precise and objective diagrams. In carrying on this type of research we have been struck increasingly by the following fact: what seem to be physiologically the same *general* kinds of species differences turn up in genus after genus and in family after family. In the flowering plants, for instance, we can now generalize even more precisely. Any two species of one genus of flowering plants generally recognized as such by competent taxonomists, may be expected to differ in all of the following ways:

epidermal cell patterns,
pubescence (size, shape, texture, distribution),
flower color pattern (white or light colored species will reveal profound differences when outcrossed with colored species),
internode pattern,
vegetative and sexual branching patterns,
petals, sepals, and leaves, will differ as follows: somewhat in size and shape, still more characteristically in trend of shape with size (i.e., the slope of the regression of width on length).

It seems significant that few if any of these differences are generally characteristic of generic differences in the flowering plants. At the present level of investigation this pronouncement is little more than an informed opinion. With a little more understanding of the morphological problem and the psychological one, it should be possible to set up definitive experiments.

Summary

Species and generic concepts as they exist in the minds of systematists were studied experimentally. It is concluded that in the simplest cases there is close agreement even between specialists who have dealt with different floras and have come to the problem with different kinds of training. It is suggested that the bases for these judgments, apparently intuitive and certainly largely inarticulate, are fit subjects for further fundamental research on the species problem.

References

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Gleason, H. A. 1952. The new Britton and Brown Illustrated Flora. 3 v. New York.

National Science Foundation

The Division of Biological and Medical Sciences of the National Science Foundation announces that the next closing date for receipt of research proposals in the life sciences is September 15, 1957. Proposals received prior to that date will be reviewed at the Fall meetings of the Foundation's Advisory panels and disposition will be made approximately four months following the closing date. Proposals received after the September 15, 1957 closing date will be reviewed following the Winter closing date of January 15, 1958.

In addition to funds for the support of basic research in the life sciences, limited funds will be available during Fiscal Year 1958 (July 1, 1957–June 30, 1958) for the support of research facilities and programs at biological field stations.

Inquiries should be addressed to National Science Foundation, Washington, D. C.

The X International Congress of Genetics

The X International Congress of Genetics will be held at McGill University, Montreal, Canada, from August 20–27, inclusive, 1958. Congress Committees are now actively preparing for your arrival and raising funds for travel grants, particularly for overseas geneticists.

The programme will include symposia, invited papers, short contributed papers, demonstrations, and other meetings and exhibits. There will be a special exhibit on "The Role of Genetics in the Service of Man." Also we are preparing an interesting programme for the ladies and children and the latter will be cared for in a "Progeny Park."

Pre- and post-Congress tours both local and extending through eastern parts of Canada and the United States are being arranged. University residences, hotels, tourist homes or camping facilities can be arranged for members and their families.

Overseas transportation should be arranged very soon as travel accommodation is already being heavily booked for 1958. Family travel plans should be investigated.

If there is any possibility that you will be able to attend, please write immediately to:

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